

Lecture 8: Nutrient Uptake

Nutrient Uptake Theory

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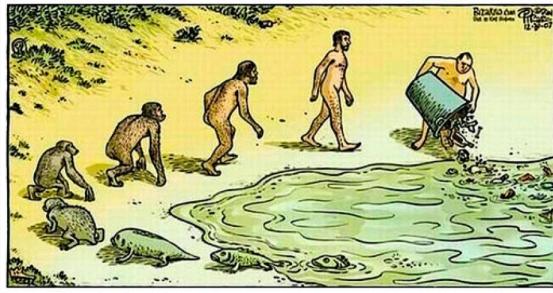
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Nitrogen: acquisition and supply



1 Introduction: The Centrality of Nitrogen

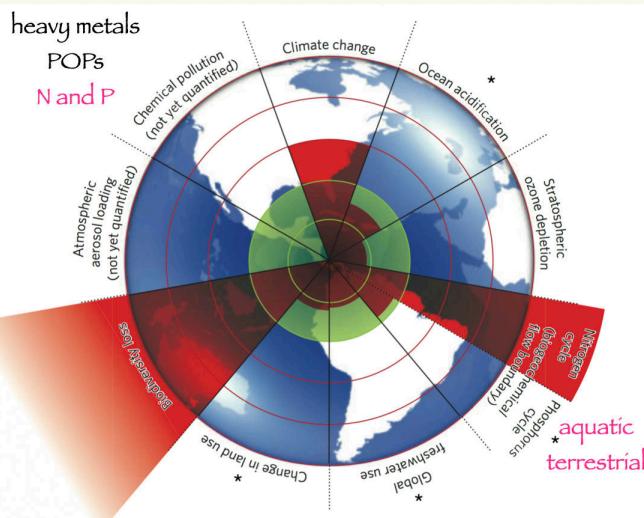


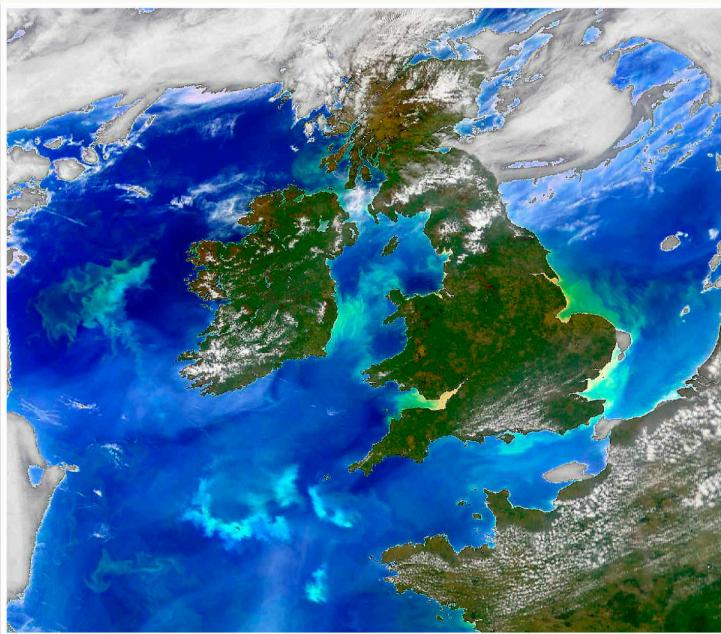
Figure 1 | Beyond the boundary. The inner green shading represents the proposed safe operating space for nine planetary systems. The red wedges represent an estimate of the current position for each variable. The boundaries in three systems (rate of biodiversity loss, climate change and human interference with the nitrogen cycle), have already been exceeded.

Today's lecture is centred on the topic of nutrient uptake. We are using nitrogen as our principal example, given its status as a ubiquitous nutrient, essential to all plants for successful growth. Additionally, we'll be considering the environmental consequences of there being excessive nitrogen in the environment. This is tied directly to the planetary boundaries concept expounded by Johan Rockström, specifically the quadrant concerning the nitrogen and phosphorus cycles—two key

global biogeochemical cycles involving the transportation and transformation of these elements between the biosphere, geosphere, atmosphere, and oceans.

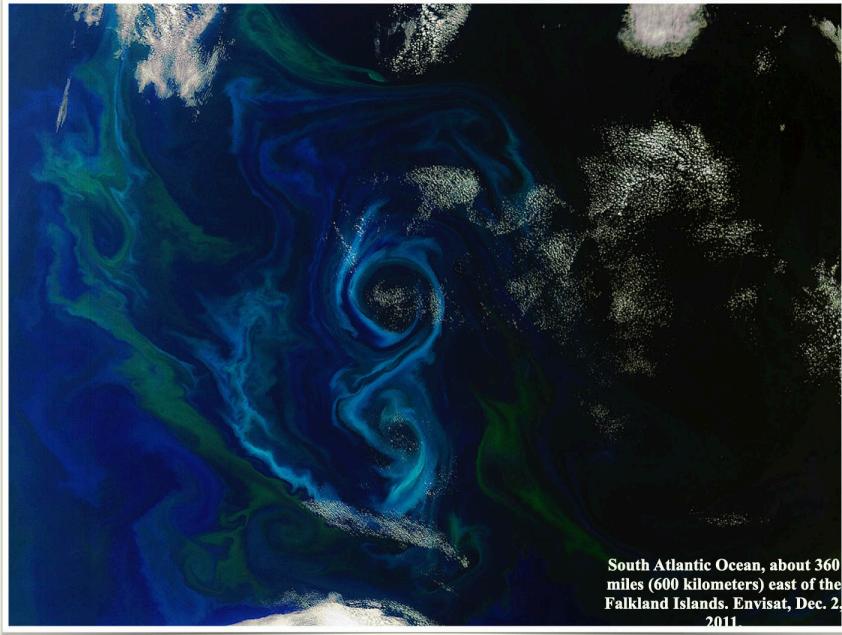
Nitrogen is a particular concern, as it is one of the major thresholds humanity has already exceeded globally. This excess results in numerous environmental problems, especially where processes involve plants—primarily aquatic and marine plants, although to a lesser extent, it does impact certain terrestrial plants as well.

1.1 Nitrogen in the Environment



Nitrogen's importance is clear when you consider its abundant presence. Approximately 79% of the air we breathe is composed of nitrogen, and it's found in soils, sand, and oceans, where it is accessed by plant roots or available in dissolved form for algae and marine plants. The most productive patches of green—on land and visible as blooms in the ocean—are areas of high nitrogen availability, where lush plant and algal growth is possible.

For this module, whilst terrestrial plants will feature in our discussions, our focus throughout the examples will be on aquatic environments, with particular attention to nitrogen dissolved in seawater and its role in triggering phytoplankton blooms. These can be so prolific that they're visible from space—swirls and green patches near the UK, Ireland, or east of the Falkland Islands, all testify to high concentrations of phytoplankton supported by nitrogen availability.



South Atlantic Ocean, about 360 miles (600 kilometers) east of the Falkland Islands. Envisat, Dec. 2, 2011.

The twirling and swirling patterns you observe in satellite imagery arise from physical ocean mixing processes—currents, eddies—distributing dissolved nitrogen, which in turn supports phytoplankton blooms.

1.2 Environmental Consequences of Excess Nitrogen



As I've mentioned, excess nitrogen in the environment, from pollution, sewage, or runoff from fertilisers, contributes to unsightly and sometimes malodorous nuisance algal blooms. These blooms

are ecologically damaging, reduce water quality, and negatively affect ecosystems and human livelihoods. They're typically accompanied by visible pond scum, floating litter, and other environmental degradation.



For instance, in China, the large population density and the dispersal of untreated sewage directly into water bodies has led to massive blooms of phytoplankton. Later in the lecture, we'll discuss the process of eutrophication, which explains in detail how these blooms develop.

Additionally, certain bacteria, such as photosynthetic cyanobacteria (“blue-green algae”), are part of the problem. As blooms expand, they block light, darkening the water and, through their respiration (especially at night), use up oxygen. Upon death, bacteria decompose the overwhelming biomass, a process which consumes even more oxygen and releases large amounts of CO₂. The result—known as a dystrophic or anoxic event—is hypoxia or complete anoxia, which leads to further die-offs, especially of aquatic animals requiring oxygen. The largest consumer of oxygen here is the decomposition of dead organic material by bacteria through respiratory processes.

2 Understanding Nutrients and Their Uptake

2.1 What Makes a Nutrient Essential?

Nutrients

Algae, like terrestrial plants, require nutrients for their **persistence**, **growth** and **reproduction**.

Algae are bathed in a nutrient medium (most of the time for intertidal species)...

No need for elaborate root systems - **uptake is through the thallus surface...** (think: links with SA:V)

Whereas many terrestrial plants have evolved symbiotic relationships with fungi or bacteria (i.e., mycorrhiza, root nodules) or specialised root systems, macroalgae have specialised nutrient uptake mechanisms to cope in low nutrient environments...

Nutrients—alongside light, oxygen, and carbon dioxide—are indispensable for plants and algae to grow, reproduce, and persist. In aquatic environments, algae are fully immersed in nutrient-rich water, allowing them to absorb dissolved nutrients directly. In contrast, terrestrial plants can only access nutrients through roots that penetrate soil, extracting dissolved nutrients from soil water.

Algae, because of their immersion, do not require roots. Their entire body (the “thallus”) is bathed in nutrients. By contrast, plants depend on root systems both for nutrient uptake and for transport to other parts of the organism. You should recall from previous modules how the surface area to volume ratio becomes decisive for nutrient uptake efficiency, particularly in aquatic environments where mixing is driven by environmental processes.

2.1.1 Symbiotic Relationships

Terrestrial plants often benefit from symbiotic relationships with fungi and bacteria—mycorrhizae and root nodules—helping them acquire and process nutrients from the soil. Aquatic algae generally do not require such associations, although bacteria in the marine environment do help make nitrogen available for algal uptake.

Bacteria, in terms of both biomass and individual numbers, are among the planet’s most abundant organisms; without them, no form of life would exist.

3 “Taxonomies” of Nutrients

Nutrients can be classified in various ways.

3.1 Essential vs Beneficial

Nutrients

Much of the knowledge of algal nutrition we have today comes from research into algal culture media between the 1930s to 1970s.

These studies have identified a host of **essential** and **beneficial** nutrients.

Essential: According to E. Epstein (1972) it is essential if

- (1) the plant can't complete a normal life cycle without it; or
- (2) the element is part of some essential plant constituent or metabolite; e.g. Mg in chlorophyll-*a*, Cl required for oxidation of water during photosynthesis

Our current understanding of plant nutrient uptake is largely indebted to studies conducted between the 1930s and 1970s. Algae, because of their direct exposure to dissolved nutrients, provided a simple and convenient model to study the principles of nutrient uptake, eventually informing our understanding of the entire plant kingdom.

3.2 A Classification Based on Physiological Effect

Nutrients

An alternative view of when a nutrient is ‘essential’:

- (i) The **alga fails to grow** or reproduce when the nutrient is limiting... **same as in Epstein's definition**;
- (ii) They **cannot be replaced** by another nutrient, i.e., no other nutrient has the same metabolic function as the limiting nutrient;
- (ii) They have a **direct effect** rather than act in conjunction with another nutrient or through some interaction with another ‘factor’.

3.3 Classification Informed by Nutrient Quantities

Nutrients

Essential nutrients classified into micro- and macro-quantities (hence **micro-** and **macronutrients**).

Reflects the relative concentrations in tissue or required in nutrient solutions (Table 5.2, Lobban and Harrison (1997); Table 4.4, Hopkins and Hüner, 2008); does not infer importance relative to the nutritional needs.

Macronutrients: mainly (but not always) structural, but some may be regulatory (e.g., Ca and Mg).

Micronutrients: catalytic and regulatory.

- **Essential versus beneficial nutrients:** Essential nutrients are those without which a plant cannot survive or complete its life cycle. Even the absence of a single essential nutrient will halt growth, productivity, or reproduction. Beneficial nutrients enhance or facilitate physiological processes, but are not strictly required for survival or completion of the life cycle.
 - By Epstein's (1972) definition, a nutrient is essential if the plant cannot complete a normal life cycle without it, and the element forms part of an essential plant constituent (e.g., magnesium in chlorophyll a).
 - Essential nutrients cannot be substituted by another element and must have a direct effect, not just act as a cofactor.
- **Macronutrients versus micronutrients:** This classification reflects the relative quantity needed by the plant. Macronutrients are present and required in much higher concentrations; their roles are often structural, contributing to the biomass of the plant (e.g., carbon, nitrogen, phosphorus, oxygen, potassium). Micronutrients, though required in far smaller amounts, function mainly as catalysts or regulators (e.g., iron in nitrate reductase).

4 So, Which Nutrients Are There?

Higher plants and algae share many (most) of the same nutrients, but their concentrations differ. Also, they differ extensively in the types of macromolecules they possess (e.g., secondary metabolites and so on), which causes the list of nutrients and their abundances to differ somewhat between these taxa.

Nutrients

About 20 nutrients fulfil critical metabolic pathways in algae.

[see Table 5.1, Lobban and Harrison (1997) for essential nutrients and their functions]

Table 5.1. *Functions and compounds of the essential elements in seaweeds*

Element	Probable functions	Examples of compounds
Nitrogen	Major metabolic importance in compounds	Amino acids, purines, pyrimidines, amino sugars, amines
Phosphorus	Structural, energy transfer	ATP, GTP, etc., nucleic acids, phospholipids, coenzymes (including coenzyme A), phosphoenolpyruvate
Potassium	Osmotic regulation, pH control, protein conformation and stability	Probably occurs predominantly in the ionic form
Calcium	Structural, enzyme activation, cofactor in ion transport	Calcium alginate, calcium carbonate
Magnesium	Photosynthetic pigments, enzyme activation, cofactor in ion transport, ribosome stability	Chlorophyll
Sulfur	Active groups in enzymes and coenzymes, structural	Methionine, cystine, glutathione, agar, carrageenan, sulfolipids, coenzyme A
Iron	Active groups in porphyrin molecules and enzymes	Ferredoxin, cytochromes, nitrate reductase, nitrite reductase, catalase
Manganese	Electron transport in photosystem II, maintenance of chloroplast membrane structure	
Copper	Electron transport in photosynthesis, enzymes	Plastocyanin, amine oxidase
Zinc	Enzymes, ribosome structure(?)	Carbonic anhydrase
Molybdenum	Nitrate reduction, ion absorption	Nitrate reductase
Sodium	Enzyme activation, water balance	Nitrate reductase
Chlorine	Photosystem II, secondary metabolites	Violaceine
Boron	Regulation of carbon utilization(?), ribosome structure(?)	
Cobalt	Component of vitamin B ₁₂	B ₁₂
Bromine ^a	{ Toxicity of antibiotic compounds(?)	{ Wide range of halogenated compounds, especially in Rhodophyceae
Iodine ^a		

^aPossibly an essential element in some seaweeds.

Source: DeBoer (1981), with permission of Blackwell Scientific Publications.

TABLE 4.4 The essential nutrient elements of higher plants and their concentrations considered adequate for normal growth.

Element	Chemical Symbol	Available Form	Concentration in Dry Matter (mmol/kg)
<i>Macronutrients</i>			
Hydrogen	H	H ₂ O	60,000
Carbon	C	CO ₂	40,000
Oxygen	O	O ₂ , CO ₂	30,000
Nitrogen	N	NO ₃ ⁻ , NH ₄ ⁺	1,000
Potassium	K	K ⁺	250
Calcium	Ca	Ca ²⁺	125
Magnesium	Mg	Mg ²⁺	80
Phosphorous	P	HPO ₄ ²⁻ , HPO ₄ ²⁻	60
Sulfur	S	SO ₄ ²⁻	30
<i>Micronutrients</i>			
Chlorine	Cl	Cl ⁻	3.0
Boron	B	BO ₃ ³⁻	2.0
Iron	Fe	Fe ²⁺ , Fe ³⁺	2.0
Manganese	Mn	Mn ²⁺	1.0
Zinc	Zn	Zn ²⁺	0.3
Copper	Cu	Cu ²⁺	0.1
Nickel	Ni	Ni ²⁺	0.05
Molybdenum	Mo	Mo ₄ ²⁻	0.001

To reiterate: macronutrients contribute to the structure and mass of the plant; micronutrients act as catalytic or regulatory substances. For example, if you removed all the iron from a large tree, you'd be left with only a handful, but that tiny amount is indispensable for the plant's metabolic processes. The table above shows the seaweed macromolecules that they contribute towards.

Algae (seaweeds) require around 20 varieties of nutrients, including:

- **Nitrogen**
- **Phosphorus**
- **Potassium**
- **Calcium**, among others.

Nitrogen is crucial—found in amino acids, nucleic acids, proteins—because proteins require nitrogen for their formation. Phosphorus is critical for structural and metabolic functions such as nucleic acids, phospholipids in membranes, and ATP transfer. Potassium and others play similar roles.

Nutrients

Unlike macronutrients (C, H, O, N and P; these also make up the bulk of the algal dry matter), most other essential micronutrients (and vitamins) are present in much smaller amounts in nature and rarely limit algal growth to the extent that the macronutrients do.

Most nutrients required for algal growth are present at much lower concentrations in the external medium (seawater) compared to in the algal thallus or cell. For example, N and P are concentrated by about 100,000 times by the seaweed, while C is about 10,000 times more abundant with respect to the medium.

In many cases this indicates some sort of **active uptake mechanism** (the nutrient is moved against a concentration gradient, a process that requires metabolic energy expenditure). More on this later.

[Table 5.2, Lobban and Harrison (1997)]

While the precise list of essential nutrients varies modestly between algae and higher plants (the latter require 17 essential elements), the principle remains the same: the majority of biomass is composed of macronutrients.

❓ Assessment Relevance

I will not set examination questions that require simple regurgitation of lists (such as “List five essential elements in seaweeds”). Focus, rather, is on understanding the processes and underlying principles.

Besides inorganic nutrients (the “bare elements,” not bound in organic molecules), plants can, through mixotrophy, also absorb dissolved organic compounds, though this is much less common and less of a focus for today’s discussion.

5 Concentration Gradients and Uptake Mechanisms

Tables commonly show, per kg of dry plant material, that macronutrient concentrations are several orders of magnitude greater than those of micronutrients.

Table 5.2 Concentrations of some essential elements in seawater and in seaweeds					
Element	Mean concentration in seawater		Concentration in dry matter		Ratio of concentration in seawater to concentration in tissue
	(mmol kg ⁻¹)	(µg g ⁻¹)	Mean (µg g ⁻¹)	Range (µg g ⁻¹)	
<i>Macronutrients</i>					
H	105,000	10,500	49,500	22,000–72,000	2.1×10^9
Mg	53.2	1,293	7,300	1,900–66,000	1.8×10^{-1}
S	28.2	904	19,400	4,500–8,200	4.7×10^{-2}
K	10.2	399	41,100	30,000–82,000	1.0×10^{-2}
Ca	10.3	413	14,300	2,000–360,000	2.9×10^{-2}
C	2.3	27.6 ^{a,b}	274,000	140,000–460,000	1.0×10^{-4}
B	0.42	4.50	184	15–910	2.4×10^{-2}
N	0.03	0.420 ^{a,c}	23,000	500–65,000	2.1×10^{-5}
P	0.002	0.071	2,800	300–12,000	2.4×10^{-5}
<i>Micronutrients</i>					
Zn	6×10^{-6}	0.0004 ^a	90	2–680	4.4×10^{-5}
Fe	1×10^{-6}	0.00006 ^a	300	90–1,500	1.0×10^{-5}
Cu	4×10^{-6}	0.0002 ^a	15	0.6–80	1.7×10^{-4}
Mn	0.5×10^{-6}	0.00003 ^a	50	4–240	2.0×10^{-5}

^aConsiderable variation occurs in seawater (Bruland 1983).
^bDissolved inorganic carbon.
^cCombined nitrogen (dissolved organic and inorganic).
Source: DeBoer (1981), including concentrations of elements in seawater from Bruland (1983), with permission of Blackwell Scientific Publications.

Figure

1:

Nutrients

In addition to the micro- and macronutrients, **vitamins** are also required by many algae and other plants. The main vitamins are B12, thiamine and biotin which are often added to the many types of nutrients solutions used for experimental purposes to promote algal growth.

More recent studies have shown that algae are also capable of utilising **dissolved organic compounds** such as amino acids (source of N) and acetic acid (source of C) as additional sources of nutrients.

A key physiological contradiction prompts interesting questions: inside the plant, the concentration of key nutrients is typically much higher than outside—in seawater or soil. For example, iron, a particularly scarce element in seawater, is maintained at relatively high tissue concentrations, which demands an energetic uptake strategy (the same applies to most other minerals and nutrients). Passive uptake via diffusion or osmosis cannot account for this, as both processes follow concentration gradients (from areas of high to low concentration).

Therefore, nutrient uptake often requires *active* transport—energy-dependent mechanisms that move nutrients against their concentration gradient into the plant, where they are assimilated into new biomass.

6 Limiting Nutrients: The Concept and Experiments



Nutrients

Early studies have identified **N as the primary nutrient limiting algal growth** in the marine environment (Dugdale, 1976).

The addition of N to seaweed aquaculture systems greatly enhances growth and production.

In some systems P is also limiting, and its addition can lead to similar growth enhancements as N additions.

Beginning in the 1960s-70s, researchers recognised that at any time, a particular nutrient could be 'limiting'. That is, if it is removed or absent, growth ceases. Professor Dugdale and colleagues demonstrated that in most seawaters, nitrogen is the major limiting nutrient. Experiments adding nitrogen to seawater samples resulted in rapid phytoplankton growth; adding other nutrients like phosphorus or potassium generally produced no such effect unless these were limiting.

Therefore, a nutrient is 'limiting' in a given context if its addition results in increased growth; if not, it isn't currently limiting. In most marine environments, nitrogen is limiting; phosphorus sometimes is, but this is more common in freshwater environments.

7 The Redfield Ratio and Nutrient Limitation

Nutrients

The Redfield ratio and nutrient limitation

Microalgae and seaweeds differ in terms of the absolute requirements of C, N and P.

In microalgae, the C:N:P ratio is approximately 106:16:1, while in macroalgae it is 550:30:1.

Macroalgae therefore require about half the P needed by microalgae, and the large amount of C indicates that macroalgae have the capacity to store or use large amounts of C (either as storage polysaccharides or used for structural support).

The 106:16:1 ratio in microalgae is called the **Redfield ratio** which has special significance to biological oceanographers.



Nutrients

N and P at a ratio of roughly 30:1 is required for optimal macroalgal growth, although in reality the range is not fixed but fairly variable:

10:1 is the optimal ratio to cultivate the commercially important red seaweed *Gracilaria gracilis*, but other studies have shown the optimal ratio to be higher at up to 80:1 for certain other seaweeds.

The optimal ratio can be determined experimentally, and this information is essential when it comes to working out fertilising strategies for seaweeds cultivated commercially in land-based tank systems. The knowledge is also used to determine a species' response to eutrophication in the natural environment as it can be used to predict the consequence of pollution.

The “Redfield ratio” is a critical empirical observation: for every 106 atoms of carbon in microalgae, there must be 16 atoms of nitrogen and 1 atom of phosphorus for optimal growth. That is, the ideal C : N : P ratio is 106 : 16 : 1.

For macroalgae, a similar ratio exists: 550 : 30 : 1 (C:N:P), reflecting the greater carbon requirement for structural integrity in larger, multicellular algae.

This optimal ratio is essential. Any deviation means one of the nutrients becomes limiting, restricting growth. Microalgae, being unicellular and minute, need less structural carbon than macroalgae.

8 Liebig's Law of the Minimum

Nutrients

Liebig's Law of the Minimum

Justus von Liebig (1803-1873; the ‘Father of the Fertiliser Industry’) published the Law of the Minimum which states that if one nutrient is deficient or lacking, algal growth will be poor even when all other elements are present at optimal or high concentrations.

In fact, the entire ecosystem productivity is limited by the nutrient that is depleted first.

Any deficiency of a nutrient, no matter how small an amount is lacking, will limit growth. If the deficient element is supplied, growth will increase up to the point where the supply of that element is no longer limiting.

Increasing the supply beyond this point is not helpful, as some other element would then be in a minimum supply and become the limiting factor.

Nutrients

The concentration of nutrients in the seawater is determined by the **balance of nutrient requirement by the alga and the rate at which nutrients can be supplied** to the alga.

The law of the minimum may also include factors (including some physical features of the environment and genetic characteristics intrinsic to the alga) that have been shown to influence algal growth.

Factors such as light intensity (affects the photosystems and the ability of the alga to take up and reduce nitrate), temperature (control the rates at which nutrients are taken up and assimilated), water motion (directly affects the ability of algae to acquire nutrients by affecting the boundary layer diffusion), algal density (enhances competition for nutrients and increases self shading, which leads to a host of follow-on effects), plant population and genetic capacities varieties and so forth may also impinge on the ability of algae to use nutrients.

Liebig's law, or “the law of the minimum,” was articulated in the 19th century. It states that the yield of a plant is determined by the single most limiting nutrient, regardless of the abundance of

others. Thus, if any one nutrient is below its critical threshold, it will restrict growth, no matter how abundantly everything else is supplied.

This principle is vital for optimising fertilisation strategies in both agriculture and aquaculture: knowing which nutrient is limiting allows for targeted supplementation for maximal growth.

Nutrients

Assume the optimal N:P ratio in the seaweed tissue is 30:1.

If N and P are present in seawater at exactly that ratio, the seaweed would grow optimally (at least for a given set of environmental conditions).

When the ratio in the external medium is >30:1?

e.g. 60:1...

growth will be limited by P

When the ratio is <30:1?

growth will be limited by N.

For a red macroalga with an optimal N:P ratio of 30 : 1, suppose more nitrogen is present than phosphorus as required. Phosphorus becomes the limiting nutrient, constraining growth despite surplus nitrogen. Conversely, if nitrogen is below the required ratio, then it is the limiting nutrient.

This concept extends to all primary producers—seaweeds, microalgae, and terrestrial plants.

9 Luxury Consumption



Luxury consumption

Any excess N or P will either **remain in the culture medium or seawater, or be taken up by the alga and stored as unassimilated inorganic N or P in the vacuoles** (in the short term), or **assimilated into some organic compound** (typically amino acids or some kind of protein in the case of N) but not used for growth.

When assimilated and stored in such a way, it may be kept internally (in the seaweed's tissue) until the limiting nutrient again becomes available when the stored nutrient is mobilised and reallocated to somatic or reproductive growth.

This principle where a nutrient is taken up and **stored in excess of immediate requirements for growth** is called 'luxury consumption', and is used as a survival strategy by many algae in unpredictable nutrient environments, or by macroalgal aquaculturists as a certain fertilising strategy.

Luxury consumption describes the phenomenon where some plants — particularly certain seaweeds — can take up more of a nutrient than is immediately required for growth, storing the excess for future use. When environmental levels of nitrogen or phosphorus later dip below optimal, the plant draws on these internal reserves to maintain growth.

This adaptation is especially valuable in environments with fluctuating nutrient availability and features prominently in aquaculture. Here, seaweeds can be provided with nitrogen and phosphorus at optimal ratios, and their ability to undertake luxury consumption helps buffer against subsequent scarcity.

Luxury consumption is a survival strategy most evident among K-selected, climax species in the ocean, conferring resilience in the face of unpredictable nutrient supply, and ensuring continued survival and growth despite external variability.

10 Environmental Consequences of Nutrients and Uptake by Seaweeds

Yesterday we spoke about nutrients. I gave you a brief introduction to what nutrients are, and explained that they can be classified into macronutrients and micronutrients, as well as essential and beneficial nutrients. Today, we need to talk about the consequences—the environmental consequences—of nutrients. We'll also begin to explore the field of measuring the uptake of nutrients by seaweeds. That's our plan for today.

Nutrients

Most of the discussion in the remainder of the lecture will use N as an example to demonstrate interesting and important points of algal nutrition.

This is because it is N that most often limits the growth of algae, but also because N pollution has negative environmental perturbations (**eutrophication**).

The theory can equally be applied to other nutrients, as the processes that govern uptake (and in many cases assimilation) are basically the same as for N.

Most concepts are equally applicable to macroalgae and microalgae.

One of the things we're going to do is to use nitrogen as our example. Nitrogen is convenient and easy to work with. The uptake mechanisms seen in many other nutrients are similar to those for nitrogen, so we can use it as a nice case study. But, of course, nitrogen is also one of the most important nutrients, both in the ocean and on land. It's often a limiting nutrient in the ocean and is important in many environmental problems we face today, such as eutrophication.

In today's lecture, I'll provide some of the ecophysiological background for why some seaweeds respond particularly well under eutrophic conditions. You will understand the physiological basis for why some seaweeds become what we refer to as nuisance algae.

10.1 Case Study: The Beijing Olympics and Eutrophication

 Invasive nature

- Beijing olympics:
Qingdao green tide



Niche models of invasive seaweed species — Heroen Verbruggen — VLIZ 2009

As an example, I'll refer to what happened during the Beijing Olympics in around 2008. Just prior to the Olympics, vast parts of the Chinese shoreline were covered with nuisance green seaweeds. Authorities had to employ a whole group of people to clean up the shoreline. All those green bits—the seaweed blooms in China—were a direct result of nitrogen entering the ocean and polluting the waterways. It's unsightly, it's smelly, and it's dangerous, so it's a huge problem around the world.

10.2 Sources and Forms of Nitrogen

Source of N

Sources of nitrogen

Nitrogen is brought into the oceans via the atmosphere (as gas) and rivers (mostly nitrates and some ammonium); it may also be lost from the ocean as nitrous oxides (NO_x) and gaseous nitrogen (N_2)

Recycling of nitrogen in the oceans is of major importance

N_2 is only used by **cyanobacteria**, but nitrate (and nitrite) and ammonium is used by most photoautotrophs in the oceans

Nutrient	Relative
gaseous N_2	95%
NO_3^-	~5 %
NO_2^-	~0 %
NH_4^+	~0.1 %

DIN:
 $\text{NO}_3^- + \text{NO}_2^- + \text{NH}_4^+$

So, where does nitrogen come from? Nitrogen is quite abundant in the atmosphere—actually, the bulk of the atmosphere is comprised of nitrogen, about 79% of it is gaseous nitrogen, N_2 . Gaseous nitrogen itself cannot be used by plants, so certain processes are required to convert that nitrogen into a bioavailable form that plants can take up.

Nitrogen is brought into the oceans via rivers, mostly in the form of nitrate and ammonium. It can also be present in the atmosphere as nitrous oxides and, in water, as nitrous oxides, entering the ocean via river runoff or various atmospheric processes. Other sources include processes in the Earth's crust, like volcanic eruptions, as well as fossil fuel burning from industrial operations, which both put nitrous oxides into the atmosphere. In certain cases, that nitrogen becomes available as a very acidic form—nitric acid—which is a source of some acid rain.

But once the nitrogen enters the ocean, many interesting processes take place. It gets recycled, taken up by algae, released by algae, released by animals that feed upon the algae—so the whole big global biogeochemical process is seen in the ocean, as well as elsewhere on the planet.

We'll delve a bit more into the detail of the global biogeochemical cycles shortly.

10.3 Nitrogen Fixation and Cycling

Cycling

P, N, Si in the oceans

Nutrient uptake by algae (phytoplankton) occurs in the euphotic zone where photosynthesis occurs. Nutrients are removed from the euphotic zone and transferred to the deeper ocean as dead organisms (detritus) sink to the ocean floor. Here, organic matter is remineralised, i.e., brought back into solution as nutrients. This process requires oxygen.

The ocean cannot support highly productive ecosystems except where nutrients are returned to the euphotic zone from below (upwelling).

Nutrient concentrations usually increase with depth, while oxygen concentration decreases. Departures from this trend are caused by advection of different water masses.

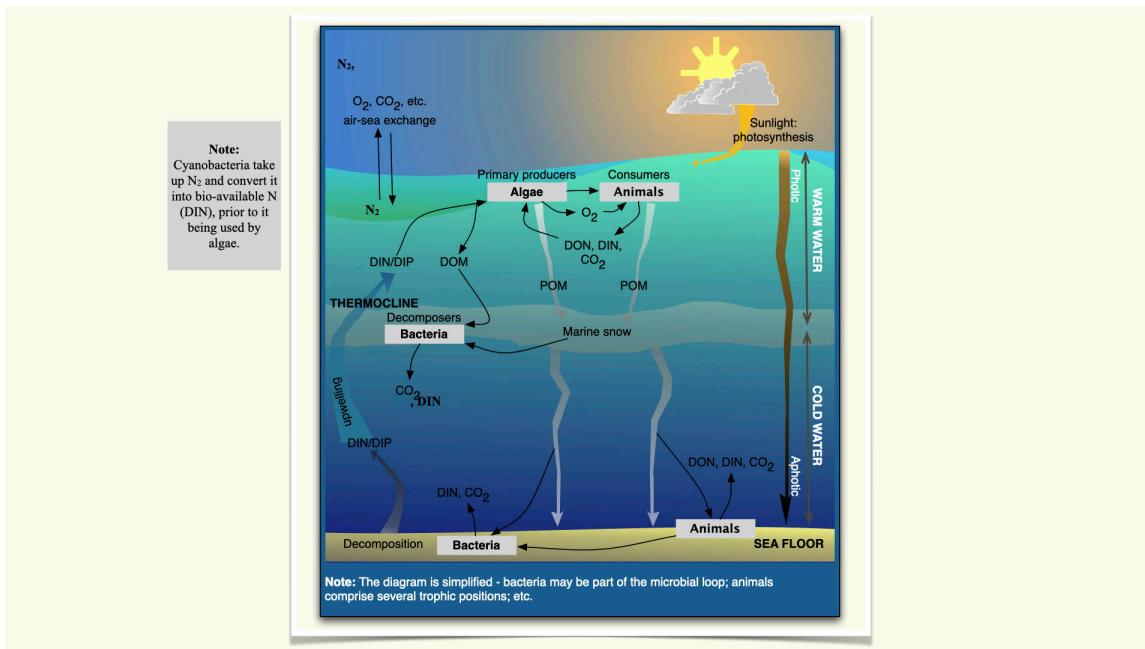
As I said before, gaseous nitrogen in the atmosphere, which is the bulk of it, cannot be used directly by plants. It must somehow become available, and this is accomplished by the process of nitrogen fixation, carried out by organisms such as cyanobacteria. Cyanobacteria can take up atmospheric dinitrogen (N_2), lock it internally in organic forms or as ammonia. When the cyanobacteria die and decompose or are eaten, that nitrogen is recycled in the form of ammonium or nitrate back into the ocean. Thus, cyanobacteria fix atmospheric nitrogen, making it available to the rest of the ecosystem to be taken up as ammonium or nitrate. This supports much of the planet's photoautotrophs, on both land and in the ocean.

If we look at the various sources of nitrogen: gaseous nitrogen is very abundant in the ocean and atmosphere. In the ocean, once it's dissolved, the total amount of all forms of nitrogen in the ocean is about 95% or so gaseous nitrogen—meaning dissolved N_2 from the atmosphere. A much smaller fraction is available as nitrate— NO_3^- —which comprises about 5% of the nitrogen in the ocean. An even smaller amount is present as nitrite NO_2^- ; it's almost immeasurable in many instances, because nitrite is only present in seawater for very short periods as an intermediary between ammonium and nitrate. Concentrations of nitrite are, therefore, very low.

Nitrogen is also present as ammonium (NH_4^+); close to about 0.1% of the total nitrogen in the ocean is present as ammonium. So those three compounds— NO_3^- , NO_2^- , and NH_4^+ (nitrate, nitrite, and ammonium)—are together known as DIN: dissolved inorganic nitrogen. This is the amount of nitrogen available for uptake by plants in the ocean.

“Dissolved” because it’s in ionic form, within the water; “inorganic” because, while it’s not bonded within an organic molecule, it does not contain carbon-hydrogen structures; and “nitrogen” because the major macronutrient atom in all these is nitrogen.

10.4 The Marine Nitrogen Cycle



Once nitrogen enters the ocean, it is cycled in various different ways. It's a complex set of reactions and processes, involving uptake by phytoplankton, their death, their consumption by zooplankton and fish, as well as decomposition—a whole host of processes.

The science that studies the transformation of various forms of nutrients between abiotic and biotic pools within the Earth system is called biogeochemistry. Biogeochemistry is concerned with the movement and the rates of transformation of nutrients between, for example, phytoplankton and zooplankton (organic or biotic components), the ocean (the hydrosphere), the atmosphere, and the geosphere.

Here is a basic, simplified representation of the nitrogen cycle in the ocean: At the top, you have the atmosphere, at the bottom the ocean floor, and in between is the water column. In and out of the atmosphere, gases such as O_2 , CO_2 , and N_2 move into the ocean, so we end up with DIN (ammonium, nitrate, nitrite) dissolved in the water. Algae then take up this DIN to produce algal biomass via photosynthesis. Animals consume phytoplankton, relying on them for biomass production, and in turn carry out respiration, taking up oxygen produced by the algae.

As animals eat algae, they excrete waste products, releasing CO_2 , more DIN, and dissolved organic forms of nitrogen into the water. In feeding on algae, animals might only consume parts, allowing algal cell contents to leak out, making dissolved organic nitrogen (DON) available to the environment. Algae can then take up this DON.

Here, you see a cycling: nitrogen comes from the atmosphere, dissolves in seawater, is taken up by algae, consumed by animals, and released again. But not all nitrogen is continually cycled—some is lost. Particulate forms of nitrogen, called POM (particulate organic matter), settle down through the water column as “marine snow.” As it falls, marine snow is decomposed by bacteria, which release

more DIN and CO_2 in the process. Thus, concentrations of DIN generally increase deeper into the ocean, as marine snow decomposes and releases more nutrients.

10.5 Remineralisation and Upwelling

Animals on the seafloor can consume marine snow, releasing DIN, DON, and CO_2 into the water column, with bacteria contributing to remineralisation. Remineralisation is the process that converts organic forms of nitrogen back into inorganic forms like ammonium and nitrate.

Over time, DIN accumulates in the deeper ocean, but physical ocean processes, such as ocean currents (upwelling), transport some of this deep, nutrient-rich water back to the surface, injecting remineralised nitrogen into sunlit upper layers, where algae can again take it up.

So, these cycles are coupled by biological processes—linking algae to animals through heterotrophy (predation, grazing), decomposition, excretion, faecal pellet production, as well as by physical processes like upwelling. Photosynthesis is a surface process, so nitrogen uptake by algae occurs mainly in surface waters, not in the deep ocean where there is no light.

Also, don't forget the role of nitrogen-fixing bacteria like cyanobacteria, which fix atmospheric nitrogen and make it bioavailable for oceanic and other organisms. This is how atmospheric nitrogen becomes available in the surface ocean.

10.6 Definitions: Some Key Terms

Cycling

In the previous slide, the basic components of the N and P cycles are:

- DIN: dissolved inorganic N
- DIP: dissolved inorganic P
- DON: dissolved organic N
- DOP: dissolved organic P
- POM: particulate organic matter

Some important definitions:

- **DIN:** Dissolved Inorganic Nitrogen; includes ammonium, nitrate, nitrite.
- **DIP:** Dissolved Inorganic Phosphorus; phosphorus equivalents to DIN.
- **DON:** Dissolved Organic Nitrogen.

- **POM:** Particulate Organic Matter; also includes particulate forms of both nitrogen and phosphorus.

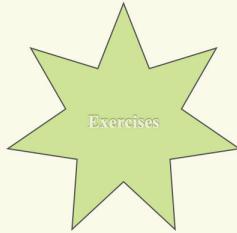
The biogeochemical cycle operates similarly on land, but most of the transformations happen within the soil, particularly around plant roots, as well as via aboveground decomposition processes—for example, as leaves fall, decompose, and transfer nitrogen back into the soil.

10.7 Units, Concentrations, and Oceanographic Patterns

Nitrogen

Units of measurement

Nutrient concentrations are usually expressed in μM units ($\mu\text{mol l}^{-1}$), but $\mu\text{g-at l}^{-1}$ may also be used.



$\text{NH}_4^+ \text{ & } \text{NO}_3^-$ ($\mu\text{mol l}^{-1}$)	$\text{NH}_4^+ \text{ & } \text{NO}_3^-$ ($\mu\text{g-at l}^{-1}$)
2	?
3	?
12	?
21	?
30	?

When reading literature about nitrogen as a macronutrient, you'll encounter various units: micromolar (μM), microgram atom per litre, and so on. These all describe the concentration of nitrogen in water or solid solution. You should recall from first-year chemistry how to convert between micromolar and microgram atom per litre (μM to $\mu\text{g atom L}^{-1}$), and vice versa. Be familiar with these conversions, as you will encounter them in tests.

$$MM = g \cdot mol^{-1}$$

$$MM = \frac{g}{mol}$$

$$\text{Molecular Mass N} = 14.0067 \text{ g} \cdot \text{mol}^{-1}$$

Have: $2 \mu\text{mol} \cdot \text{l}^{-1} \text{ NH}_4^+$

How many μg N?

$$14.0067 = \frac{\mu\text{g}}{2 \mu\text{mol}}$$

$$28.0134 \mu\text{g}$$

Note that these calculations (*i.e.* those involving macronutrients N and P) are based only of the atom of interest, *e.g.* N in this example, within the overall molecule.

The Prefixes Used with SI Units			
Prefix	Symbol	Meaning	Scientific Notation
exa-	E	1,000,000,000,000,000,000	10^{18}
peta-	P	1,000,000,000,000,000	10^{15}
tera-	T	1,000,000,000,000	10^{12}
giga-	G	1,000,000,000	10^9
mega-	M	1,000,000	10^6
kilo-	k	1,000	10^3
hecto-	h	100	10^2
deka-	da	10	10^1
—	—	1	10^0
deci-	d	0.1	10^{-1}
centi-	c	0.01	10^{-2}
milli-	m	0.001	10^{-3}
micro-	μ	0.000 001	10^{-6}
nano-	n	0.000 000 001	10^{-9}
pico-	p	0.000 000 000 001	10^{-12}
femto-	f	0.000 000 000 000 001	10^{-15}
atto-	a	0.000 000 000 000 000 001	10^{-18}

You must also know the SI prefixes and the number of zeros associated with each—grammes, milligrammes, microgrammes, nanogrammes, picogrammes, et cetera. In plant physiology, a basic grasp of chemistry and SI unit prefixes is assumed.

10.8 Typical Oceanic Nitrogen Concentrations

Nitrogen

Environmental concentrations...

In tropical areas and some temperate seas and oceans such as the temperate coastal areas of Australia, concentrations can be almost immeasurable all year round.

In coastal upwelling areas along the west coasts of all major continents (the California Current along western shores of North America; the Benguela Current along the west coast of southern Africa; the Canary Current along the west coast of north Africa; and the Humboldt Current which runs along South America) nutrient concentrations vary seasonally, with maximal nutrient concentrations (total inorganic N = 20 – 40 μM ; inorganic P = ~2 μM) during the major upwelling season and lowest concentration the rest of the time (generally <4 μM total inorganic N; <0.2 μM inorganic P).

In upwelling systems, nutrients may become available in pulses every week or two, each one lasting one to several days, depending on local oceanographic and climatic conditions.

Here's a range of concentrations that nitrogen is available in the ocean:

- **Tropical regions (ca. 10°S to 10°N):** Very low nitrogen; concentrations may be in the nano- to picogramme range (ng L^{-1} – pg L^{-1}).
- **Most of the ocean:** Microgramme to milligramme range.
- **Freshwater systems:** Often reach the milligramme range.
- **Upwelling regions (e.g., the Benguela upwelling off South Africa, Canary Current off North Africa, Humboldt off South America, California Current off North America):** Highest oceanic nutrient levels. Here, total inorganic nitrogen can reach up to 40 $\mu\text{mol L}^{-1}$, while inorganic phosphorus typically reaches 2 $\mu\text{mol L}^{-1}$.

Note the ratio here, approximately 10:1, closely corresponding to the Redfield ratio.

During active upwelling, nutrient concentrations rise to 20–40 $\mu\text{mol L}^{-1}$ nitrogen, 2 $\mu\text{mol L}^{-1}$ inorganic phosphorus. When upwelling subsides, these values drop below 4 $\mu\text{mol L}^{-1}$ for nitrate and 0.2 $\mu\text{mol L}^{-1}$ for inorganic phosphorus.

Thus, in the ocean, background nitrogen levels are not fixed but fluctuate, sometimes very rapidly (over tens of minutes) due to oceanographic processes such as upwelling. Algae have evolved various adaptations to cope with the intermittent and transient nature of nitrogen availability in the ocean —a stark contrast to terrestrial systems, where changes are usually slower.

10.9 Classification of Oceanic Systems Based on Nutrient Levels

Nitrogen

Most natural marine ecosystems fall within the two categories mentioned above – systems with year round low nutrient status are called **oligotrophic**, while the upwelling systems are called **mesotrophic**.

Eutrophic systems have unnaturally high amounts of nutrients.

Let's look at the history of eutrophic systems, and why they exist.

The ocean can be classified into:

- **Oligotrophic:** Low nutrient, e.g., tropical regions, almost no nitrogen.
- **Mesotrophic:** Intermediate, e.g., upwelling zones, fluctuates temporally.
- **Eutrophic:** High nutrients, often due to human influence—unnaturally high nitrogen.

In the ocean, nitrogen is typically limiting. However, excessive input causes problems—most notably, eutrophication.

10.10 The Nitrogen Bomb: Human Impact

Nitrogen

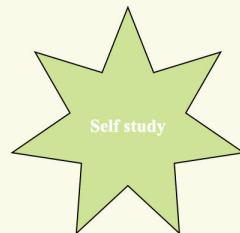
Usually N limitation is not the problem... (although in aquaculture...)

A problem of global proportions

The Haber-Bosch process has successfully short-circuited the nitrogen cycle so that the distribution of nitrogen globally was significantly modified. Today more nitrogen is fixed annually by anthropogenic processes than by natural nitrogen fixation. Modifications to the nitrogen cycle originally manifested itself in increased agricultural production — this is the desired effect and is seen as beneficial — but gradually negative effects were witnessed in aquatic and coastal marine environments.

Eutrophication has been described as the oldest problem of water quality caused by human activities in lakes and coastal ecosystems and has today become a problem of global proportions.

Nitrogen



The Nitrogen Bomb

By learning to draw fertilizer from a clear blue sky, chemists have fed the multitudes. They've also unleashed a fury as threatening as atomic energy
By David E. and Marshall Jon Fisher
Photographs by James Worell
DISCOVER Vol. 22 No. 04 | April 2001

See 'The Nitrogen Bomb (2001).pdf' on iKamva.

Also see:

The Scientist Who Killed Millions and Saved Billions
<https://www.youtube.com/watch?v=EvknN89JoWo>

For your self-study: read the article "The Nitrogen Bomb" (available on Ecoma). The Haber-Bosch process has resulted in a huge problem worldwide in both terrestrial and aquatic systems. "Nitrogen bomb" is a metaphor for the disastrous potential of excessive and unwisely applied nitrogen, most sharply observed in eutrophication. This is examinable content.

10.11 What is Eutrophication?

Eutrophication

One consequence of eutrophication in natural systems is the development of **blooms of unwanted ('nuisance') opportunistic macroalgae** such as *Ulva*, *Enteromorpha* and *Cladophora* spp. (and phytoplankton in certain systems).

Blooms are unsightly, but there is a host of other effects such as **shifts in community composition** (i.e. species diversity, abundance, and biomass), **and in severe cases anoxia and dystrophic events** due to the decomposition of algal biomass after blooms.

A system's response to eutrophication varies greatly, and depends on the scale of the nutrient inputs. The term 'bloom' used describes the situation where a system becomes dominated by a high biomass with an incredibly low diversity of species (note the similarity here with upwelling and mariculture systems).

Eutrophication usually occurs when too much nitrogen is added to a body of water that would naturally be nitrogen limited. In these systems, certain algae or seaweeds respond rapidly to the enrichment. For instance, the three illustrated species all possess high surface area to volume ratios, meaning nearly every cell is exposed directly to the environment and can immediately take up available nutrients. This capacity allows rapid growth and biomass accumulation.

Blooms

Bloom development is as much influenced by increased nutrient pools over and above that which is available naturally, as by the morphology (and hence physiology) of the algal thallus.

Refer to the Functional Form model (Littler & Littler, 1980); basically, it states that macroalgae with a large SA:V such as membranous *Ulva* spp., or finely branched species such as *Cladophora* spp. have faster rates of nutrient uptake than species with a coarser or thicker thallus construction (Wallentinus, 1984).

The higher rate of nutrient uptake of opportunistic species (with high SA:V) imparts a competitive advantage over other groups of macroalgae under eutrophic conditions (Rosenberg & Ramus, 1984).

More complex seaweeds with lower surface area to volume ratios respond more slowly, if at all, to such nutrient pulses; the response is more distributed and structurally limited. In contrast, these high surface area opportunistic algae (sometimes called R-selected species) bloom excessively when nutrients are introduced, becoming nuisance species and disrupting the ecological balance.

In a natural system, there is a high diversity of plants and animals. After eutrophication, one species becomes dominant, reducing community composition, species richness, and causing the biomass of that one species to increase exponentially.

In severe cases, the system can become anoxic. Imagine a dense bloom of photosynthesising algae; at night, without photosynthesis, respiration consumes all the oxygen in the water. Species that require oxygen die off, and their decomposition consumes even more oxygen, eventually producing low-oxygen, or dystrophic, conditions.

Bacteria are key to these processes, as they drive decomposition and thus increase total ecosystem respiration and oxygen consumption.

10.12 Mitigating Eutrophication

As for what can be done: removing the blooming nuisance algae is not addressing the underlying issue. We must ensure that sources of nitrogen entering the water are addressed—by improving sewage treatment, reducing fertiliser runoff from agriculture, and proper waste management. Rivers seen as convenient dumping grounds simply transfer the problem downstream; the consequences are borne by someone else or by the ecosystem.

11 Linking Form and Function: Surface Area to Volume Ratio

I've mentioned that response to eutrophication is connected to both the morphology and physiology of algae. Those species with high surface area to volume ratios can absorb nutrients rapidly and outcompete others. This is where Littler and Littler's "functional form model" comes into play, explaining why morphology is crucial to ecological dynamics under eutrophic conditions.

An ecophysiological understanding

We will focus on the physiological properties of nuisance macroalgae that give them a competitive advantage in eutrophic systems. In order to find out why only certain species respond to nutrient additions we will also examine the physiology of those species that do not bloom, but which persist or even thrive under oligotrophic or mesotrophic conditions.

There are also other points that may be of interest:

What is the source of nutrients to primary producers in very oligotrophic systems?

How is the physiology of seaweed nutrient uptake (uptake kinetics) studied?

In order to understand the nutrient physiology of seaweeds and the mechanisms seaweeds employ to cope with transient or more consistent nutrient environments, it is important that we first look at the basic concepts of seaweed nutrition.

In summary, you should now connect previously disconnected ideas—such as surface area to volume ratio and nutrient uptake. Understanding this enables a more comprehensive view of how eutrophication alters ecosystems.

If you have questions or do not grasp a particular aspect, you're welcome to ask on WhatsApp. Please ensure you read the assigned articles and refresh your knowledge of unit conversions and SI prefixes, as you will be expected to use this knowledge fluently.

Read further on eutrophication. Much more can be said, but the core is straightforward biology playing out in an ecosystem that simplifies under stress—one species dominates as nutrients increase, reducing diversity and altering physiological and ecological processes within the system.

12 Nutrient Uptake

Today, I want to discuss the physiology of plant nutrient uptake, specifically focussing on the processes by which algae—seaweeds, in marine environments—take up nutrients from their surroundings. Throughout this lecture, I'll be using the terms 'algae', 'seaweed', and 'plant' interchangeably; for the purposes of our discussion, they all refer to seaweeds, as the experiments we're considering were conducted with seaweeds. The uptake processes in terrestrial plant roots are similar regarding the mechanism of transfer from the environment into the plant, but the main differences lie in the environmental processes within soils.

Our goal is to develop an ecophysiological understanding of how plants (seaweeds) absorb nutrients, which will also help explain how seaweeds thrive in various marine habitats, and why many can become nuisance species under eutrophic conditions.

12.1 Pathways and Barriers to Nutrient Uptake

Entry of DIN into cells

water column/soil water → across boundary layer → across cell wall
→ across cell membrane (plasmalemma) → into cytoplasm

When nitrogen or any other nutrient is taken up, its pathway runs from the water column (or surrounding soil water), across a thin, static barrier of still water adjacent to the cell wall or membrane, known as the boundary layer. From there, nutrients cross the cell wall and membrane, entering the cytoplasm. The uptake of solutes across these solid-fluid boundaries is a key process in nutrient cycling across aquatic, marine, and terrestrial ecosystems.

Uptake kinetics

light intensity
heat
nutritional history

water motion (reduce boundary layer)
conc. gradient (incr. diffusion rate)
morphology (SA:V)

The transport of solutes across solid-fluid boundaries plays an integral role in nutrient cycling in marine and aquatic systems.

The movement of nutrients from the aqueous medium into seaweeds is described by a two stage process:

- (i) the uptake rate is influenced by the diffusion across the boundary layer, the thin layer of water adjacent to the algal surface.
- (ii) reaction kinetics inside the thallus, mediated by the seaweed itself makes up the final stage of uptake. When fast diffusion rates can be sustained, nutrient acquisition is kinetically controlled (limited); conversely, uptake of N is controlled (limited) by diffusion rates when kinetic reactions are faster than what can be supplied across the boundary layer.

Nutrient uptake can be described as a two-stage process:

1. **External Phase:** Processes taking place outside the plant cell, primarily governed by the diffusion of nutrients across the boundary layer. This thin, unmoving water layer is held close to the algal surface by hydrostatic and intermolecular forces.
2. **Internal Phase:** Once nutrients have crossed the membrane, internal processes—especially enzymatic reactions—rapidly convert inorganic nutrients into organic forms. Enzymatic reaction rates, described as reaction kinetics, control how swiftly nutrients such as ammonium or nitrate are metabolised into amino acids, proteins, and other cell constituents.

When assessing nutrient uptake, it is important to distinguish between these two phases. If diffusion from the environment is very rapid, then the overall rate is limited by how quickly the plant's enzymes can process the nutrients—in other words, the internal, kinetically controlled processes. Not all plants possess the same enzymatic processing rates; some transform nutrients quickly, others more slowly.

Conversely, if environmental diffusion is slow, the uptake rate is limited by external physical processes, rather than by the plant's internal processing capacity.

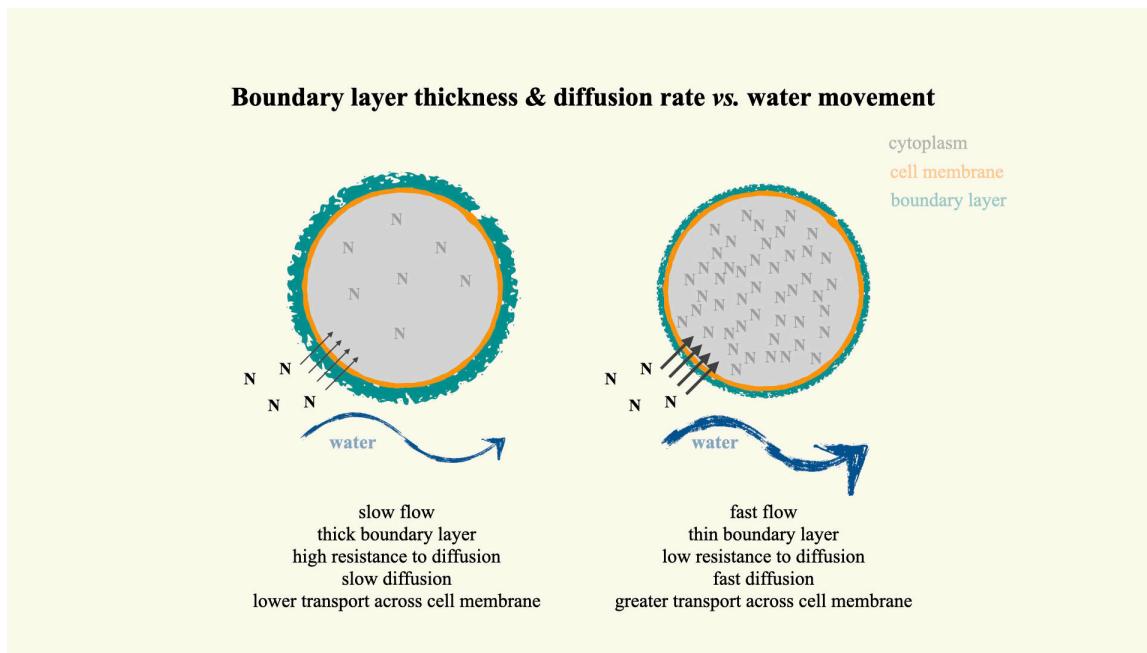
12.2 Factors Influencing Nitrogen Uptake

Nitrogen (and other macronutrient) uptake is influenced by many factors:

- **External Conditions:** The thickness of the boundary layer and the actual concentration of nutrients in the water are crucial. These are influenced by physical conditions outside the plant.
- **Form of Nutrient:** Ammonium is taken up much faster than nitrate; nitrate requires active uptake, while ammonium is acquired via passive diffusion.
- **Nutrient Starvation History:** Plants recently starved of nutrients will uptake rapidly when exposed to fresh supply; non-starved plants may not show a significant response.
- **Environmental Conditions:** High light intensity and high temperatures both enhance metabolism and, consequently, increase V_{max} , speeding up uptake rates.
- **Surface Area to Volume Ratio:** As discussed, this has a substantial effect on uptake dynamics.
- **Mechanisms of Uptake:** The presence of additional mechanisms (such as facilitated diffusion) can also influence overall nutrient uptake.

All of these together influence the rate at which macronutrients such as nitrogen and phosphorus are taken up from the environment.

12.3 The Boundary Layer Concept

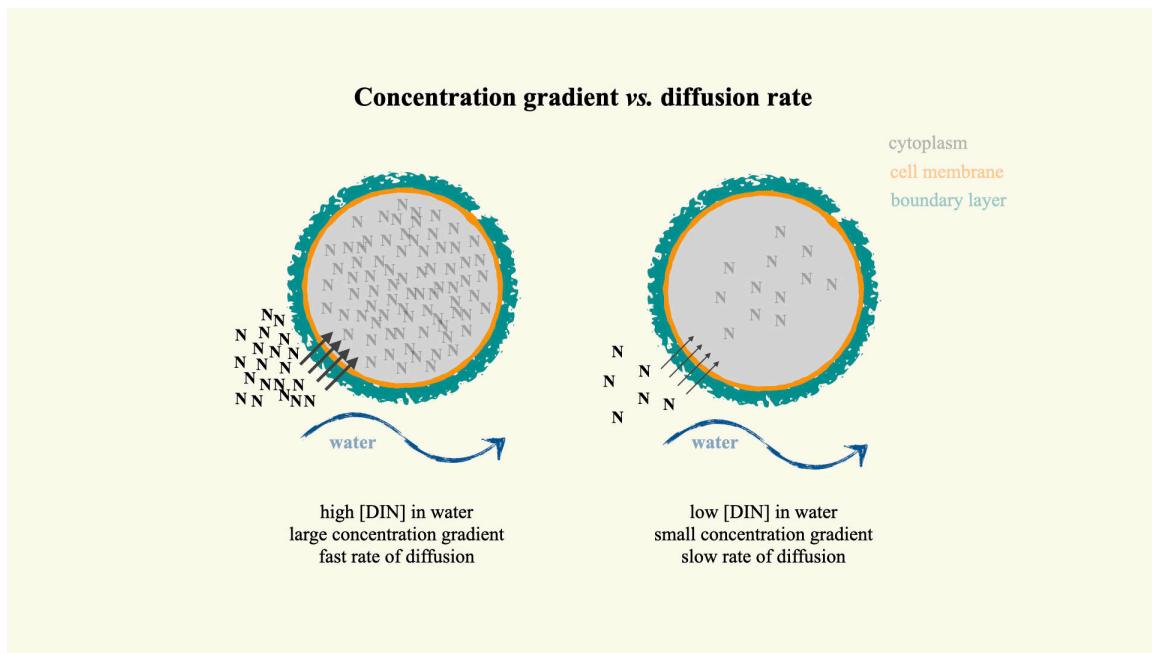


Let me illustrate the boundary layer effect. Picture a plant cell in still water: the cell membrane (orange in our diagram) is surrounded by the boundary layer—a static skin of water held onto the surface. The thickness of this layer is influenced by water movement: in still conditions, it is thick; with greater water motion, it becomes thinner.

The boundary layer acts as a resistance barrier to diffusion. The thicker it is, the greater the distance and resistance for molecules diffusing from the environment into the cell, thus lowering the flux (rate) of nutrient transfer. Where water moves rapidly, the boundary layer thins, reducing resistance and allowing greater nutrient movement into the cell.

Thus, in turbulent waters, plants take up nutrients more efficiently; in still waters, uptake is limited due to the thick boundary layer.

12.4 Influence of Environmental Nutrient Concentration

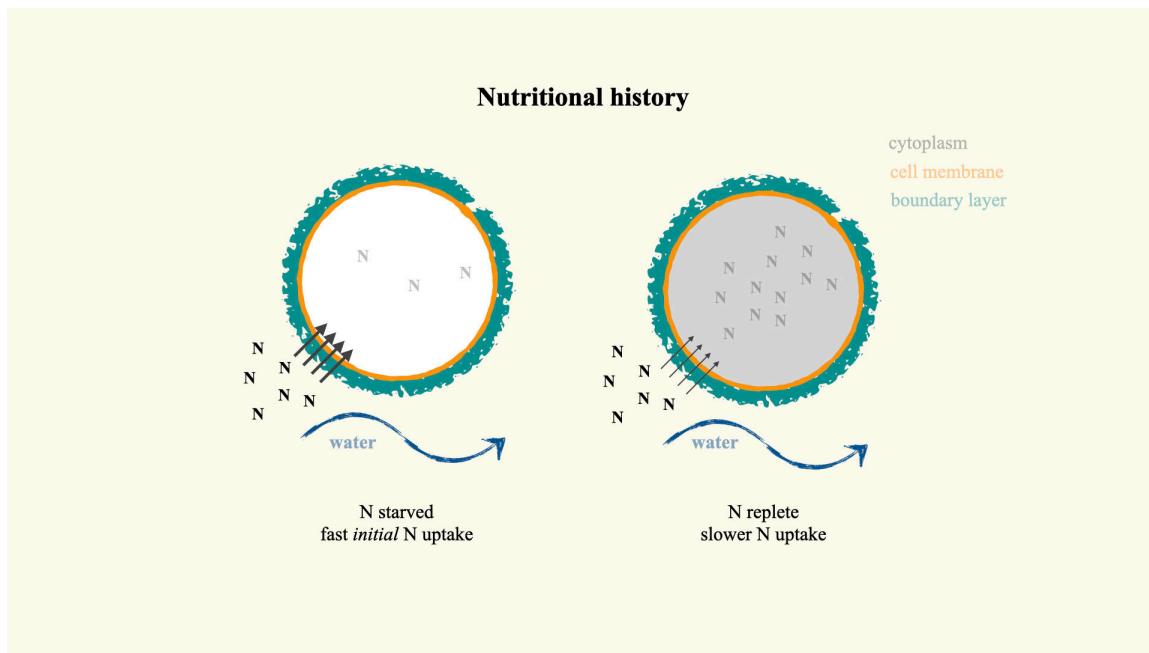


Water movement isn't the only factor affecting uptake. Another is ambient nutrient concentration. With identical water movement, a cell surrounded by a high nutrient concentration will experience a steeper concentration gradient, promoting greater nutrient flux into the cell. The rate of diffusion is directly proportional to the gradient between external and internal nutrient levels.

In very low-flow conditions, increasing the concentration of external nutrients can compensate somewhat for a thick boundary layer, enhancing uptake despite slow water movement.

These two principles—the thickness of the boundary layer (and thus water movement) and the nutrient concentration gradient—underlie the role of the diffusion step in limiting nutrient uptake.

12.5 Algal Nutritional History



Another way in which nutrient uptake is affected—this time due to properties within the interior of the plant cell or algal cell—is when you have algae that have been starved of nutrients for an extended period. For instance, in situations in the ocean characterised by periods of low nutrient availability, such as during the non-upwelling phases (downwelling) in upwelling systems, the continued growth of algae, despite the limited external supply, results in diminished concentrations of internal nutrients within the algal cells.

Consequently, when these nutrient-depleted, or nutrient-starved, cells are subsequently transferred into seawater with a high concentration of nutrients, the rate of nutrient uptake increases dramatically. If you then compare the nutrient uptake rate of these nutrient-starved cells to seaweed or algal cells that have a history of recent exposure to nutrients—in other words, where the internal nutrient concentration is much higher—you will find a marked difference. Specifically, if you compare the rate of uptake between nitrogen-starved algae and nitrogen-replete algae, the uptake rate in the starved cells will be far greater than in the nitrogen-replete cells. The reason for why this big difference exists will become clearer later on.

13 Measuring Nutrient Uptake in the Laboratory

💡 Supplement Your Study of Nutrient Uptake:

A highly detailed protocol for conducting nutrient uptake experiments is provided in “Lecture 8b: Uptake Kinetics – Michaelis-Menten.”

Uptake kinetics experiments

Please see Tangled Bank for essential additional reading:
https://tangledbank.netlify.app/BDC223/L08b-michaelis_menten.html

The whole uptake process of nitrogen by algae can be measured *in situ* or under controlled conditions in the laboratory.

Nutrient uptake is often determined by measuring the **disappearance of a nutrient from the culture medium over a time interval** after the addition of the alga (using either the ‘batch-mode’ or ‘perturbation’ approach).

Such experiments allow the calculation of **depletion curves**, and from the depletion curve the **uptake kinetics** can be determined.

To quantify nutrient uptake in plants, algae, and seaweeds, we perform controlled experiments in laboratory flasks. Here's the protocol I followed for my PhD research:

Required reading

Botanica Marina Vol. 45, 2002, pp. 196–209 © 2002 by Walter de Gruyter · Berlin · New York

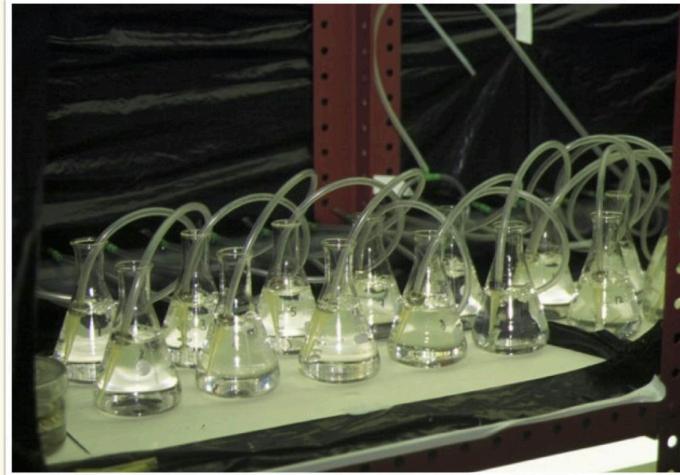
Nitrogen Uptake by *Gracilaria gracilis* (Rhodophyta): Adaptations to a Temporally Variable Nitrogen Environment

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The physiology of nitrogen acquisition was determined for *Gracilaria gracilis* (Stackhouse) Steveth, Irvine *et al.* Farham in a series of perturbation experiments with the aim of examining uptake kinetics in response to transient nutrient disappearance. Experiments demonstrated that varying environmental variables such as nutrients, NO_3^- -N and NH_4^+ -N concentrations and interactions, temperature and water motion affect parameters of linear and Michaelis-Menten models. A third ‘Michaelis-Menten parameter’ (α) is introduced here and added to the model to describe the effect of water motion on nutrient uptake. Nutrient uptake kinetics were best described by a linear, rate-unlimited response, with the slope increasing with N limitation, indicating that *Gracilaria* is more efficient at acquiring nutrients when internally stored N pools were impoverished. Temperature had a significant effect on nutrient uptake. At low temperatures (10°C), NO_3^- -N uptake was suppressed by approximately 38 % in the presence of NH_4^+ -N at concentrations above 5 μM , and the seaweed displayed a higher affinity for NH_4^+ -N than for NO_3^- -N at low temperatures. Nitrate-nitrogen uptake followed a typical Michaelis-Menten pattern, with the rate of uptake increasing with increasing NO_3^- -N concentration until it reached a plateau. Ammonium uptake was also Michaelis-Menten-like, but it did not enhance the affinity for NO_3^- -N (only in N-limited thallus), while nutrient limitation enhanced affinity irrespective of temperature. The maximal velocity of uptake (V_{max}) and the half saturation constant (K_s) appeared to vary with temperature. The results suggest that water motion may reduce diffusion limitation and thus reduce diffusion transport limitation experienced by the alga under conditions of low external dissolved inorganic nitrogen (DIN) concentrations, so that the rate of N uptake responds with a 4.5-fold increase under conditions of high water motion. The results of this study indicate that *Gracilaria gracilis* is a highly productive in an upwelling environment dominated by the transient availability of DIN through the use of a high affinity system for NO_3^- -N and non-selective uptake of NH_4^+ -N. Water motion interacts strongly with nutrient concentration, and may alleviate N limitation by reducing boundary layer resistance to diffusion. Practical application of the results of this study is discussed in terms of significance to aquaculture.

Introduction
Gracilaria gracilis (Stackhouse) Steveth, Irvine *et al.* Farham occurs as free-living, largely monospecific beds on a limited number of sheltered coastal water bodies on the west coast of southern Africa. Studies on the ecology of *Gracilaria gracilis* and *Gracilaria tikvahiae* in Namibia and South Africa have been reported by Anderson *et al.* (1980), Rottmann (1990), Molby (1990), Anderson *et al.* (1990), Anderson *et al.* (1994a, 1996a), Smit *et al.* (1997), Smit (1998), Anderson *et al.* (1998, 1999) and Smit and Botha (1999). In Namibia, the growth rates of *Gracilaria* are low, environmental nutrient concentrations appear to be responsible for the low growth rates of *Gracilaria* (Anderson *et al.* 1990). In South African waters, similar seasonal changes in growth rates and production of seaweeds have also been ascribed to nutrient limitation (e.g. Huisman and Ramanayake 1982, Lapointe and Duke 1984, Fujita *et al.* 1989, Borum and Sand-Jensen 1996). It is well known that the addition of N can greatly enhance the growth rate and production of seaweeds under certain conditions (Lapointe and Ryther 1979, Smit *et al.* 1997). Anderson *et al.* (1996a) also suggest that site-related differences in growth rates of *Gracilaria* may be explained by differences in water movement at these sites.

The growth rate and productivity of algae is in part, controlled by the availability of dissolved inorganic nitrogen (DIN) in the aqueous medium surrounding the thallus (Dugdale 1967, Chapman and Craigie 1977, Chapman and Craigie 1980, Chapman 1984, Chapman 1991). The ability of an alga to utilize N for biomass production is determined by the rate at which DIN can pass through the boundary layer of the thallus, the rate at which N takes part in biochemical processes (Walter 1980, Koch 1980, Koch and Wiegert 1980, Chapman 1984). Control of internal physical processes acting on the boundary layer governs the latter of diffusion, whereas enzymatic processes within the algal thallus determine the pre-stage of uptake. Understanding N nutrition of seaweeds thus requires the integration of at least three controlling factors, i.e. N concentration in the growth



Depletion curves

Depletion curves display the **decrease in concentration over a period of time** from when an algal sample is placed into a nutrient solution until the end of the experiment when all/most nutrients had been taken up.

Depletion curves allow us to ask the question, “**How much N does a unit of seaweed take up in a unit of time?**”

- A convenient ‘unit’ of seaweed is a gram (but it is your choice... *select something sensible*).
- A convenient ‘unit’ of time is an hour (but it is your choice... *pick something sensible*).
- “How much N” generally implies “how many μg of N,” but you may also work in μmol units...
- Note, the unit ‘ μM ’ is not appropriate when asking “how much N?”

Each flask contains seawater with a known, enriched nutrient concentration and a measured piece of seaweed. At both the start and end of a timed interval (usually 20 minutes), water samples are taken to measure nutrient concentration. The decrease in nutrients reflects uptake by the seaweed.

We also introduce bubbling into the flasks to facilitate water movement, thereby thinning the boundary layer and promoting diffusion. Without bubbling, the boundary layer thickens, reducing nutrient uptake.

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- “How much N” generally implies “how many µg of N,” but you may also work in µmol units...
- Note, the unit ‘µM’ is not appropriate when asking “how much N?”

Something that we need to keep in mind when we design these uptake experiments, and when we calculate depletion curves, is that we need to be very certain of the units of measurement that we use during the conduction of our experiment. Typically, we would want to select a certain amount of seaweed—the mass of seaweed that we are going to place within a certain volume of water—and we are going to measure the rate of disappearance of that nutrient from that water over a certain amount of time. We also have to have an idea of the amount of nutrients present in the water.

Firstly, when we work with seaweeds, a typical mass unit of measurement would be grams. Most of the seaweeds that we can work with are of a size where we can weigh them and represent their mass in a couple of grams to tens of grams. Some of the larger seaweeds would weigh in the order of kilograms, and so on. So, we need to be sure that we use a unit of measurement that is appropriate for the thing being studied.

Nutrient uptake as a process can happen very quickly. It can happen over the space of minutes. Certainly, within a period of less than an hour, we can very easily measure the rate of disappearance of nutrients from the culture medium. So, a convenient unit for time measurement would be per hour. But again, depending on the organism that you are going to study, you will have to pick something sensible and relevant to that organism.

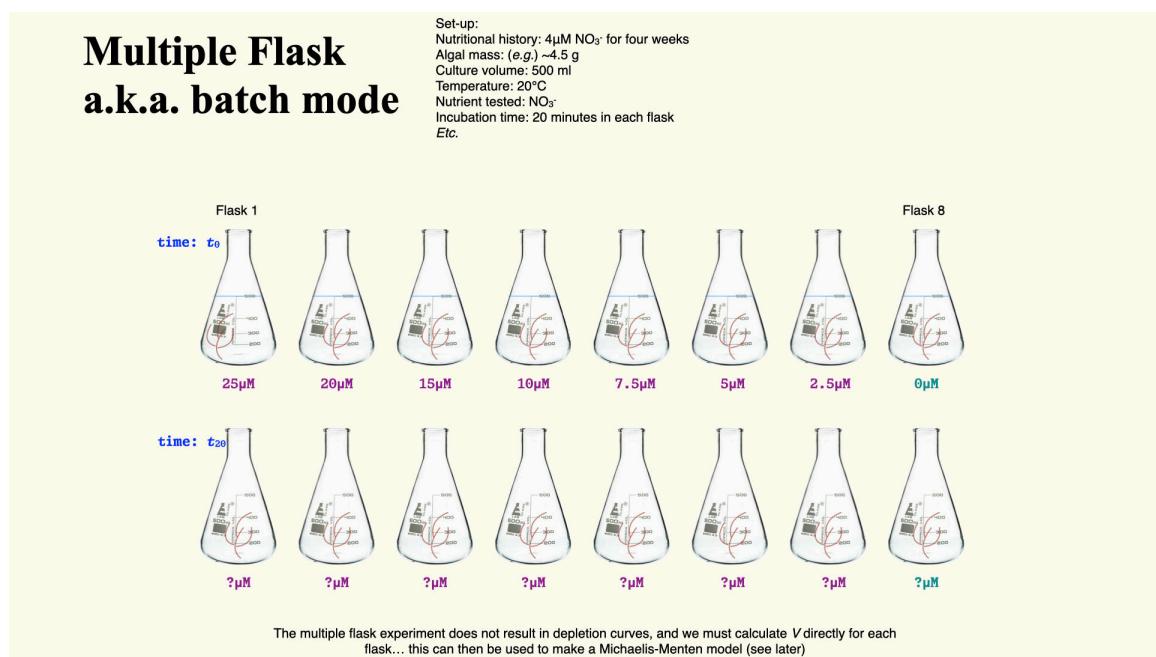
When we talk about the volume of the culture medium (usually dictated by the size of our experimental containers), typically we are going to take a small piece of seaweed, algal tissue excised from the whole plant, and put it into a smaller volume of water. That volume of water is often something that can very easily fit into a flask. So, the units of measurement there would be in the millilitre range. Often, the amount of seawater contained in this experiment is, say, less than a litre. In my experiments, it was 250mL or so, if I remember correctly.

Then, concentration units. There are different ways that we can represent chemical concentrations—that is, the concentration of the nutrient in question. We can measure concentrations in micromolar, for example $\mu\text{mol L}^{-1}$ (μM), or we can represent concentrations in micrograms per litre, $\mu\text{g L}^{-1}$. Again, be sensible in terms of what unit you choose to represent your measurements in.

It is very important that we know these units right at the beginning of our experiments, because they are going to carry through multiple stages of calculations, so that we represent an uptake rate in a sensible unit such as for moles of nutrients taken up per gram of seaweed per hour, i.e., $\text{mol g}^{-1}\text{h}^{-1}$. For example, we normalise data to a ‘unit’ of seaweed—typically, per gram, since different flasks might contain slightly different masses (e.g., 1.2 g versus 1.1 g). By dividing the uptake rate by available mass, we standardise uptake on a per-gram basis. Similarly, we standardise to unit of time by converting measurements taken over 20 minutes to an hourly rate by simple multiplication. Remember, from past lectures, conversion between units (micrograms and micromolar, for instance) is sometimes required.

13.1 Types of Uptake Experiments

13.1.1 Multiple flask experiment



In a multiple flask experiment, each flask begins with a different initial concentration of nutrient (e.g., $25\text{ }\mu\text{mol L}^{-1}$, $20\text{ }\mu\text{mol L}^{-1}$, $15\text{ }\mu\text{mol L}^{-1}$, etc.), the same amount of seaweed, same water volume (e.g., 500 mL), and the same temperature (e.g., 20°C). After a set duration, changes in nutrient concentration indicate how much has been taken up. Calculations are carefully outlined for you and involve correcting for flask volume, time, and seaweed mass.

You will notice in the figure above that we have a range of concentrations going all the way from about $25\mu\text{M}$ down to $0\mu\text{M}$ concentrations. Why is it that we have $0\mu\text{M}$? Obviously, no nutrients can be

taken up when we have no nutrients present. So, the answer to that question is fairly straightforward: it is a control.

A control is always necessary in our experiment to establish that the only process influencing the mechanism we are studying is the subject of the study—in this case, the nutrients themselves. Therefore, in the $0\mu\text{M}$ flask, what we would expect to observe after 20min—when we remeasure the amount of remaining nutrients in the solution—is that it is still going to be $0\mu\text{M}$.

There are some processes that occur in seawater that can put nutrients back into the water. By having a flask set at $0\mu\text{M}$ nitrogen, we can verify that none of those processes are present within our experiment. In other words, we are controlling for any process that may reintroduce nutrients into the water. If we discover that there is a process that does put nutrients back into the water, we can determine the magnitude of this effect and subtract that from the values we establish for each of the experimental conditions in which nutrients are actually present.

13.1.2 Calculating N uptake in multiple flask experiments

The process of calculating nutrient uptake rates from the depletion curves, which we can establish from the multiple flask experiments, might at first appear complicated. There are indeed many steps involved and multiple points along the way where it is necessary to translate units from one form to another. However, if you understand the process thoroughly, all of these steps become extremely logical and systematic.

Calculations (e.g. flask 1)

$t_0: 25\mu\text{M } \text{NO}_3\text{-N}$
 $t_{20}: 9.9\mu\text{M } \text{NO}_3\text{-N}$
Algal mass: (e.g.) 4.5 g
Culture volume: 500 ml

Repeat for every other flask

Calculating V from the multiple flask data

Step 1: How much N taken up in 20 minutes?
 $> 25\mu\text{M} - 9.9\mu\text{M} = 15.1\mu\text{M}$... this is the reduction in the N concentration but it says nothing about how much *i.e. the mass N was removed* ...
So, let's work with the mass of N instead.

Step 2: Convert concentrations to mass N present per flask at the start and end.
Knowing that $\mu\text{M} = \mu\text{mol.L}^{-1}$, how many μg N is $25\mu\text{mol.L}^{-1}$, and how many μg N is $9.9\mu\text{mol.L}^{-1}$?
 $\text{MM} = \mu\text{g}.\mu\text{mol}^{-1}$, and the MM of N is 14.0067g.mol^{-1} , therefore...
 $> 14.0067 = \text{x}\mu\text{g} / 25\mu\text{mol} = 350.17\mu\text{g N}$ and
 $> 14.0067 = \text{x}\mu\text{g} / 9.9\mu\text{mol} = 138.67\mu\text{g N}$
This is the mass of N in 1 L at the start and end ... *but we have only 500 mL in the culture flask!*
So, what mass of N in 500mL?
So, initially we had ...
 $> 350.17\mu\text{g N} / 2 = 175.09\mu\text{g N}$
... and after 20 minutes we had ...
 $> 138.67\mu\text{g N} / 2 = 69.34\mu\text{g N}$

Step 3: How much N does the 4.5g alga take up in 20 minutes?
 $> 175.09\mu\text{g N} - 69.34\mu\text{g N} = 105.75\mu\text{g N}$

Step 4: How much N does 1 g alga take up in 20 minutes?
 $> 105.75\mu\text{g N} / 4.5\text{g} = 23.5\mu\text{g N.g}^{-1}$

Step 5: If $23.5\mu\text{g N.g}^{-1}$ is taken up in 20 minutes, how much in 1 hr?
 $> 23.5\mu\text{g N.g}^{-1} \times 3 = 70.50\mu\text{g N.g}^{-1.\text{hr}^{-1}}$

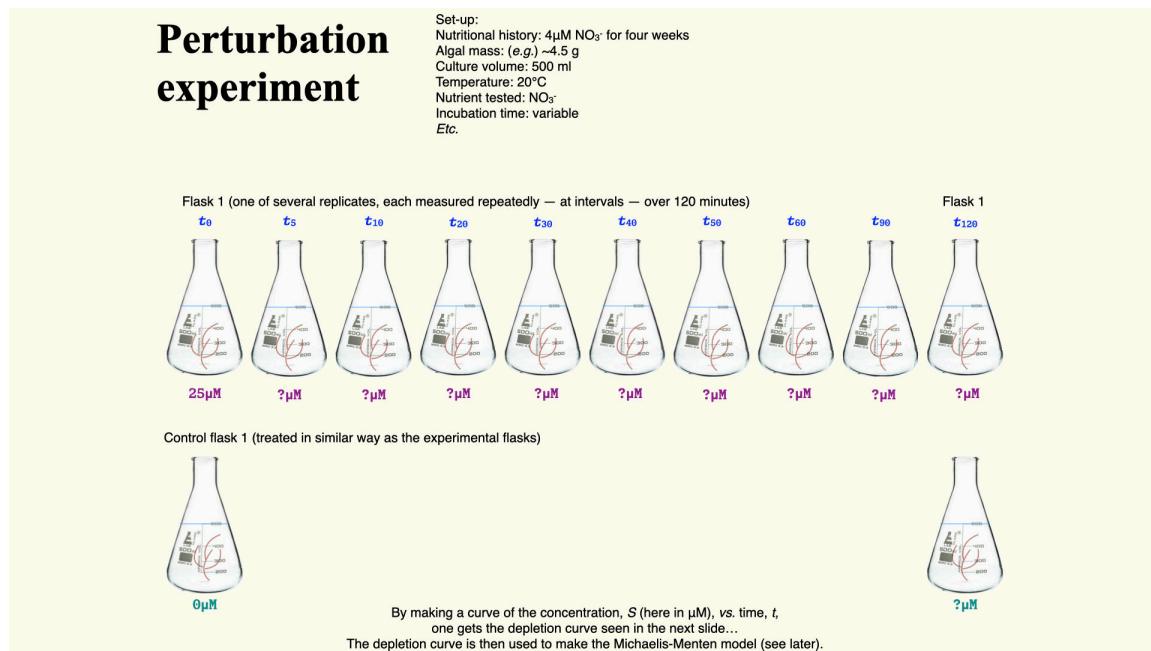
It becomes quite straightforward with sufficient knowledge—of course, practical experience helps tremendously—but at the fundamental level, if I simply provide you with the mass of seaweed used in our experiment, the exact duration for which they are exposed to the experimental conditions, and the concentrations of nutrients in the various flasks both before we begin the experiment and, let's say, after 20 minutes, you should be able to work things out. If I give you only those values, you

can, by applying a logical system of calculations, arrive at the required answer, that is, the rate of nutrient uptake.

The answer to this seemingly complex question is reported in the units you see in the final result: the amount of nutrient taken up per gram of seaweed per hour, or ($\frac{\Delta \text{ units of nutrient}}{\text{g seaweed} \cdot \text{h}}$). Simply by knowing what your final units are—which, in this case, is a rate—you have already laid out all the key steps needed to get from the start of the experiment to your final output.

In fact, the method for calculation can often be deduced just by carefully considering the unit of measurement in which we report the result. If you break it down, the logical operations to get to that unit will show you each calculation required, provided you start with sound, well-measured data.

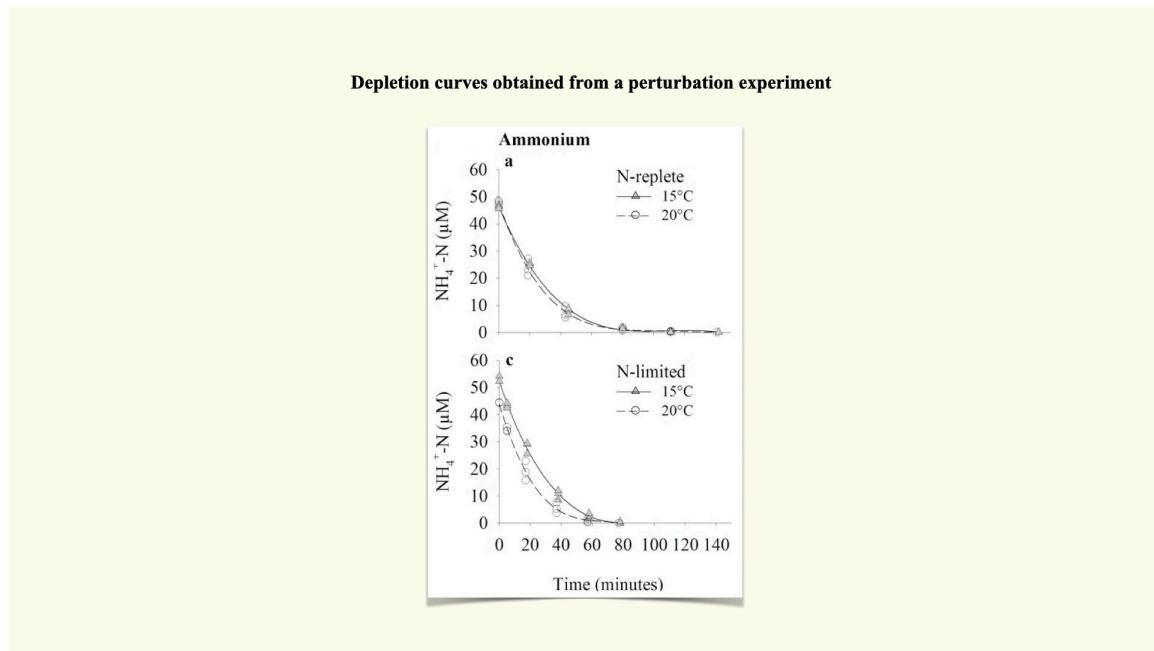
13.1.3 Perturbation experiments



With the perturbation experiment (the approach used in your spreadsheet data you will encounter in the Lab), a single flask is set up with, for example, 25 $\mu\text{mol L}^{-1}$ nitrate and a known mass of seaweed. At regular intervals (e.g., every five minutes), water samples are removed and nutrient content analysed. Rather than resetting the experiment for each time step, you use the same flask, tracking nutrient decline over time until it is depleted. The control flask should show no change, confirming the validity of the experiment.

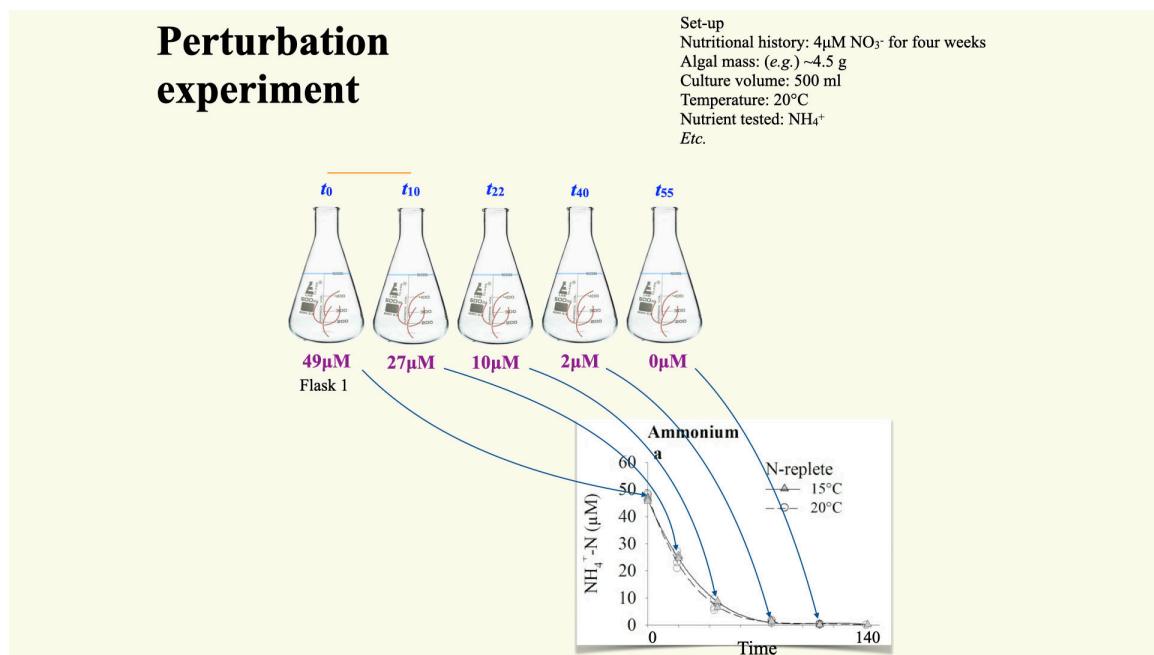
This generates a depletion curve: plotting nutrient concentration over time, often for several replicate flasks at differing environmental conditions (e.g., high/low temperatures, pre-starved or already nutrient-replete seaweeds).

13.1.4 Calculating depletion curves from perturbation experiments

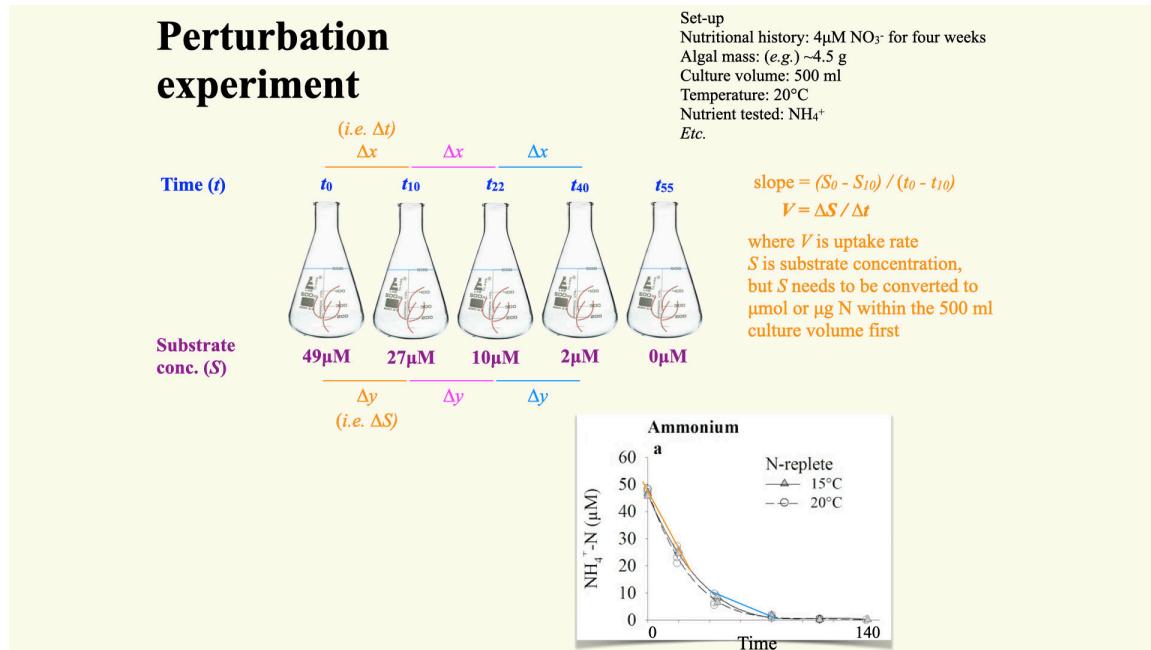


The uptake rate, denoted V , is the rate of nutrient uptake per gram per hour (or other unit of time). This is clearly shown in the depletion curves. Mathematically, it is the change in substrate concentration, ΔY , over change in time, ΔX :

$$V = \frac{\Delta Y}{\Delta X}$$



As you can see in the slide above, plotting a depletion curve is very straightforward. All you need to do is recognise that time is the independent variable; that is, it will be plotted on the x -axis. The nutrient concentrations remaining in the flasks after each consecutive time interval are plotted on the y -axis, which serves as the dependent variable. Once you have constructed the plot as I have just described, you will observe a curve very similar to the one I have shown for ammonium uptake.



For each time interval, you calculate this rate from the depletion curve. At the start—where the external nutrient concentration is highest—uptake rate is fastest, and the curve (shown above by the straight line fitted to the interval, whose slope is V) is steepest. As the nutrient is depleted, the curve flattens and V decreases.

Here is the sequence of steps, which are as logical and systematic as in the multiple flask experiments:

Calculations (e.g. first time interval, $t_0 - t_1$)

t_0 : 25 μM $\text{NO}_3\text{-N}$
 t_1 : 21.3 μM $\text{NO}_3\text{-N}$
 Algal mass: (e.g.) 4.5 g
 Culture volume: 500 ml

Repeat for every consecutive time interval, i.e.
 $t_{10} - t_9$
 then
 $t_{20} - t_{19}$
 etc.

Calculating V from the depletion curves

Step 1: How much N taken up in 5 minutes?
 $> 25\mu\text{M} - 21.3\mu\text{M} = 3.7\mu\text{M}$... this is the reduction in the N concentration but it says nothing about how much (i.e. the mass) N was removed ...
 So, let's work with the mass of N instead.

Step 2: Convert concentrations to mass N present per flask at the start and end.
 Knowing that $\mu\text{M} = \mu\text{mol.L}^{-1}$, how many μg N is 25 $\mu\text{mol.L}^{-1}$, and how many μg N is 21.3 $\mu\text{mol.L}^{-1}$?
 $\text{MM} = \mu\text{g}.\mu\text{mol}^{-1}$, and the MM of N is 14.0067 $\text{g}.\text{mol}^{-1}$, therefore...
 $> 14.0067 = x\mu\text{g} / 25\mu\text{mol} = 350.17\mu\text{g}$ N
 and
 $> 14.0067 = x\mu\text{g} / 21.3\mu\text{mol} = 298.34\mu\text{g}$ N
 This is the mass of N in 1 L at the start and end ... but we have only 500 mL!
 So, what mass of N in 500mL?
 So, initially we had ...
 $> 350.17\mu\text{g N} / 2 = 175.09\mu\text{g N}$
 ... and after 20 minutes we had ...
 $> 298.34\mu\text{g N} / 2 = 149.17\mu\text{g N}$

Step 3: How much N does the 4.5g alga take up in 5 minutes?
 $> 175.09\mu\text{g N} - 149.17\mu\text{g N} = 25.92\mu\text{g N}$

Step 4: How much N does 1 g alga take up in 5 minutes?
 $> 25.92\mu\text{g N} / 4.5g = 6.48\mu\text{g N.g}^{-1}$

Step 5: If 5.76 $\mu\text{g N.g}^{-1}$ is taken up in 5 minutes, how much in 1 hr?
 $> 6.48\mu\text{g N.g}^{-1} \times 12 = 77.76\mu\text{g N.g}^{-1}.\text{hr}^{-1}$

These rates (V) are then used for the next phase of analysis.

14 Different Uptake Mechanisms

Uptake mechanisms

Uptake may be of one of several types: (1) **active**; (2) **passive transport**; (3) **facilitated diffusion** transport; and 4) **biphasic uptake**.

Before we dive deeper into the calculation of uptake kinetics—in other words, kinetics meaning the study of the rate processes underlying some biological process—I think it is useful to distinguish between a couple of different ways in which seaweeds can take up nutrients.

There are four different kinds of nutrient uptake mechanisms. For the immediate sections following below, we will be talking about **active uptake**. In other words, this is a form of uptake where nutrient acquisition is driven by the expenditure of energy. This is typically the kind of uptake process that we study when we conduct multiple flask or perturbation experiments.

Another type of uptake—the second type—that we will cover later on in the lecture, which can also be studied by constructing depletion curves, is the process of **passive transport** or **passive uptake**. This is where nutrient uptake is entirely driven by processes involving the diffusion, the diffusive flux, of nutrients into the cells.

Much later on, we will talk, but not in too much detail, about **facilitated diffusion transport** and **biphasic uptake**.

For the next few sections in our lecture that continue below, the perturbation and multiple flask methods that we are applying will be showcasing how we would go about studying active transport. In other words, we will focus on cases where energy is expended to take up nutrients, and where the flux of these nutrients can proceed against the concentration gradient.

15 Uptake Kinetics: Michaelis-Menten Model

Uptake kinetics: V vs. S

(i.e. uptake rate as a function of substrate concentration)

Uptake rate (V) for the **active mechanism** is often described as a hyperbolic function of **substrate concentration (S)**, by analogy to the Michaelis-Menten expression used to model enzyme catalysed reactions.

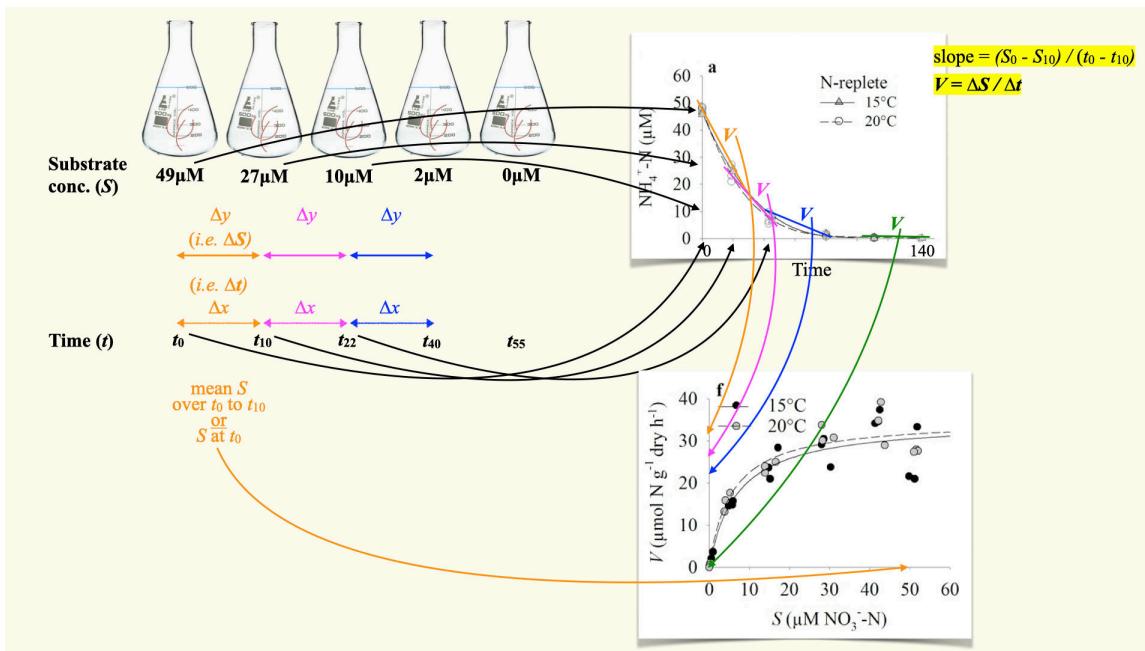
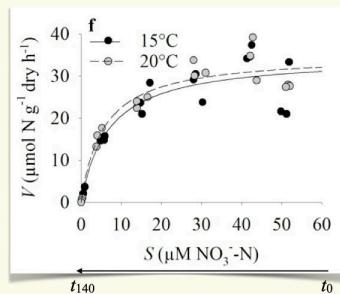
The Michaelis-Menten equation assumes that uptake is unidirectional so that no losses occur after uptake.

We then relate uptake rate (V) to the substrate concentration (S) we typically do so by generating a V vs. S plot. This relationship follows the Michaelis-Menten equation, familiar from enzyme kinetics, resulting in a characteristic hyperbolic curve:

- At high substrate concentrations (initially in the experiment), uptake rate is maximal and governed by the enzymatic processing capacity—the internal kinetic limit.
- As substrate is depleted, the rate drops off, and diffusion through the boundary layer becomes limiting—the external physical limit.

Michaelis-Menten

$$V = V_{\max} \cdot \frac{S}{K_s + S}$$



When plotting V against S , we see:

- A plateau at high external concentrations, where processes are enzyme-limited, called V_{max} . It is important that, if seen as a time-related process, V_{max} occurs at the **start of our experiment** (in perturbation experiments), or as seen from the point of view of a multiple flask experiment, at the highest nutrient concentrations. In both instances, this is the slope of the line segments fitted to the **start of our depletion curves**.

- A curved downturn at lower concentrations, where diffusion and transport limitation take over, which is the process captured by the **tail ends** of the depletion curves (**end of the process**).

$$V = \frac{V_{\max} \cdot S}{K_s + S}$$

Here, V_{\max} is the maximum uptake rate (set by enzyme capacity), K_s is the half-saturation constant (substrate concentration at which V is half V_{\max}), and S is substrate concentration.

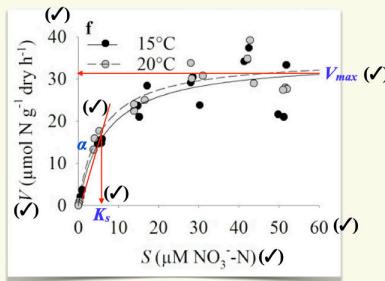
When a graph of uptake rate (V) versus substrate concentration (S) displays a hyperbolic tangent curve—a steep rise at low concentrations and a plateau at high concentrations—this signifies **active** or **facilitated uptake**. The maximum rate of uptake (V_{\max}) is primarily set by factors intrinsic to the algae, such as the enzymatic processes involved.

Consequently, environmental factors that influence enzyme activity—like light intensity or temperature—will impact how high V_{\max} can be. Thus, active uptake is largely determined and influenced by environmental conditions that promote growth, photosynthesis, and metabolic activity—for example, high temperatures and abundant light.

15.1 Reading the Michaelis-Menten Graph

Michaelis-Menten

V_{\max} , which is the (extrapolated or theoretical) maximal rate of uptake of the nutrient of interest under the experimental conditions, and K_s , which the half saturation constant and is numerically equivalent to the value of S , the substrate concentration, where $V = \frac{1}{2} V_{\max}$.



At high S , uptake is only limited by enzyme processing speed—even if you double or triple nutrients, V remains at V_{\max} . Once the environmental nutrients start to diminish, the rate V declines, indicating the transition to diffusion-limited uptake.

- V_{\max} —maximal enzyme processing rate.
- K_s —substrate concentration where V is half V_{\max} , indicating the plant's ability to extract nutrients at low concentrations.

- α (alpha)—slope of the initial portion of the curve, reflecting affinity for nutrients in low-nutrient environments.

A low K_s (or steep α) indicates high affinity, making certain algae better adapted to low-nutrient conditions. A high K_s suggests poor adaptation to nutrient poverty.

15.2 Applications and Importance

Active uptake

The parameters V_{max} and K_s have an **ecological meaning** since they describe the nutrient uptake ability of a species under specific environmental conditions and allows for comparisons of nutrient uptake kinetics among species and studies.

Understanding these parameters helps explain why some seaweeds become opportunistic, blooming in nutrient-rich environments while others persist under nutrient limitation. The values of V_{max} , K_s , and α are affected by species' physiology, surface area to volume ratio, light, temperature, pre-conditioning, and other ecophysiological parameters.

You should now be able to connect plant ecophysiology and uptake models to the practical experiments and analysis required for your coursework. Remember to relate all elements back to both phases (diffusion and enzymatic reaction), and know which environmental or physiological factors are likely to limit uptake under different scenarios.

Uptake kinetics

A knowledge of the factors affecting nutrient uptake by seaweeds (and plants), and how it differ between the types nutrients of is critical to our understanding of the effects of nutrient availability on growth and production (Fujita, 1985).

Many studies have focused on the **uptake kinetics** of ammonium and nitrate but fewer have looked at the kinetics of nitrite uptake because of its low natural concentration (it is quickly converted from ammonium to nitrate via the action of two types of bacteria, *i.e.*, *Nitrosomas* and *Nitrobacter*; additionally, is toxic at high concentrations, as is ammonium).

Right, so today we will continue to talk about nutrient uptake. Last week, we spoke about nutrient uptake experiments, and I showed you how to derive information from the depletion curve—the relationship that shows uptake rate versus substrate concentration. When we plotted that relationship, the graph appeared as a hyperbolic tangent curve. At low concentrations, the uptake rate increases rapidly, and then at high nutrient concentrations, it reaches a plateau. This type of relationship is known as the Michaelis–Menten uptake relationship, and it serves as an example of one of three different uptake mechanisms called active uptake.

On Thursday, we will discuss passive transport and facilitated diffusion, which are two additional uptake mechanisms. Generally speaking, algae and most other plants can display one of these three mechanisms—active transport, passive transport, and facilitated diffusion. Today, our focus will remain on active uptake.

Last week, after we explored the uptake curve—the V versus S relationship—of active uptake as determined for nitrate, I explained the various parameters: V_{max} , K_s , and α . On this slide, you will find text discussing the ecological significance of these parameters. We have covered this before, so I will not repeat it in detail. Instead, let us delve a bit further into what active uptake involves.

16 Active Uptake

Active uptake

K_s describes the **affinity** of the carrier site to a particular nutrient: the lower K_s , the higher the affinity (high affinity: better cope under low nutrient concentrations).

The parameter α is the initial slope of the Michaelis-Menten model calculated at concentrations of less than K_s . It also indicates the affinity for a particular nutrient and the **ease at which the alga is able to respond with a faster nutrient uptake rate upon increases of the nutrient** in the external medium.

α is independent of V_{max} (while K_s is not independent of V_{max}) which makes it more useful than K_s in indicating affinity for a nutrient; it is useful for comparing the competitive abilities of different algae.

The ecological meaning attached to V_{max} , on the other hand, is that it describes **the maximal rate at which nutrients can be taken up when it is available in abundance**.

Active uptake allows most plants to maintain nutrient concentrations inside their cells that are much greater than those found in the external environment. This process enables nutrients to be transported from an area where there is a lower concentration to an area inside the plant where there is a higher concentration—essentially moving against the concentration gradient. More formally, this occurs against the electrochemical potential gradient.

Active uptake

The transfer of ions or molecules across a membrane **against an electrochemical-potential** gradient:

External concentrations: usually in the micromolar range;

Internal concentrations: usually in the millimolar range;

i.e. **passive diffusion unlikely**

To quantify, external nutrient concentrations are usually in the micromolar range, while internal concentrations inside the cell are generally in the millimolar range—that is, from $\mu\text{mol L}^{-1}$ externally to mmol L^{-1} internally. This large difference suggests that passive diffusion alone is insufficient, as diffusion would only allow movement from high to low concentration.

Passive diffusion is – at most – responsible for the movement of nutrients from the environment across the boundary layer. This specific process is determined by passive diffusion. However, the major uptake of nutrients into the cell is described by active uptake. For this to occur—from a region of low concentration to one of high concentration—cells must expend energy, namely metabolic energy.

The energy expended is generally light-dependent, with ATP being the most likely source. This is achieved by a system that involves proton-pumping ATPases, setting up a gradient between the external and internal environment in terms of pH. The proton gradient, or the pH gradient, establishes the electrochemical potential gradient, which then drives secondary ion transport. The secondary ion in question is the nutrient—such as nitrate, in our previous example.

This coupling between the pH gradient and the nutrient gradient facilitates the active uptake of nutrients. Coupled transport may arise from differing movements of ions at different sites, either in opposite or the same direction. When hydrogen ions are pumped out of the cell and nutrients are pumped in, this form of counter-transport is known as anti-port, or anti-porter transport. Conversely, some nutrients move in the same direction as the protons—this is called symport or co-transport.

In algae, the proton pump is linked to the co-transport of substances like sugars and thiourea, and there are also mechanisms involving the pumping of sodium ions, which can be responsible for the co-transport of other nutrients from the environment into the cell.

16.1 Key Points of Active Uptake

You must remember that cellular energy, primarily derived from ATP, is required to drive active uptake. ATP drives the proton pump, which establishes the primary gradient, and then nutrients are brought into the cell by coupling to this gradient.

This is the mechanism by which an energetic, active process brings nutrients into the cell—by coupling with a proton pump generated by ATP and ATPases. While this is a complex physiological process, knowing this overview will suffice; we will not explore all the fine physiological details.

16.2 Characteristics that Define Active Uptake

Active uptake

Other characteristics: **selectivity of ions; saturation of the carrier system** (e.g. Michaelis-Menten, see later)—these are also characteristic of facilitated diffusion.

Active transport is seen in most algae (but...)

Active uptake

The hyperbolic geometry of the V vs. S curve suggests that in most cases uptake is not simply a passive process relying on diffusion alone, but that it is **active** or **facilitated**, i.e.

- (i) that it is controlled to some extent by **factors intrinsic to the alga** itself; or
- (ii) it may be the case that some other **upper limit is imposed** on the rate at which nutrients can be incorporated into thallus tissue.

Beyond its energy requirement, active uptake is defined by several additional characteristics:

1. **Selectivity for Particular Ions:** Only specific ions are taken up via active transport, not all. For example, nitrate, phosphorus, and sulphates can be taken up in this way. One of the components of dissolved inorganic nitrogen (DIN), ammonium, is typically not taken up via active transport and is excluded here.

- Saturation of the Carrier System:** There is a stage in the uptake process where the carrier system becomes saturated. At high nutrient concentrations, there is a portion of the curve where V_{max} is reached—meaning uptake rate will not increase, despite increasing external nutrient concentration. This is because the enzymatic systems responsible for transport become saturated and cannot operate any faster.
- Movement Against the Concentration Gradient:** As previously discussed, active uptake involves the movement of ions against their concentration gradient.

When uptake rate (V) is plotted against substrate concentration (S) and the curve shows a steep initial rise followed by a plateau—i.e., a hyperbolic tangent shape—we can infer the process is mediated by either active or facilitated uptake. The maximum uptake rate, V_{max} , is controlled by intrinsic factors within the algae, specifically those that govern enzyme function. Thus, environmental factors that influence enzyme activity, such as light intensity and temperature, will affect V_{max} .

Put simply, environments promoting rapid growth—higher temperature, more light—will increase enzyme activities, resulting in a higher V_{max} .

These features—selectivity, saturation, and movement against the gradient—define active uptake. Note that some of these factors also apply to facilitated uptake, which we shall discuss later.

16.3 Surface Area to Volume Ratio and Uptake Parameters

Suppose we gather several seaweeds, spanning all six or seven different functional form categories outlined by Littler and Littler, and conduct uptake experiments. We would discover that V_{max} and K_s vary as a function of the surface area to volume ratio.

Plants with flat, membranous, or highly filamentous forms tend to grow rapidly and thus have a much higher V_{max} —thanks to their high surface area to volume ratio, which allows every cell direct exposure to the nutrient-rich environment. They are also typically able to acquire nutrients effectively even in environments where nutrient levels are low, implying they often have a low K_s (and thus a high affinity for nutrients).

On the other end of the spectrum, algae with a low surface area to volume ratio—where the bulk of the cells are internal—grow more slowly, with reduced access to light and slower diffusion rates. Consequently, they possess a low V_{max} and often a higher K_s , making them less able to acquire nutrients when these are scarce.

If these low V_{max} species are placed in a nutrient-rich (eutrophic) environment, it makes little difference, because their limitation is set by internal cellular processes, not external nutrient supply. In contrast, fast-growing, high-surface-area species with high V_{max} and low K_s will respond rapidly to eutrophic conditions. This helps explain why some algae become nuisance or problematic under excessive nutrient conditions—their physiological traits make them well-suited to exploit high nutrient environments.

Understanding the ecological significance of high or low V_{max} and K_s is crucial; it explains the circumstances under which species will thrive and proliferate based on the environmental nutrient regime.

16.4 The Three Phases of Active Uptake

Phases of active uptake

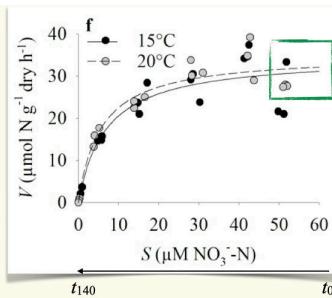
The hyperbolic model can also be discussed in terms of phases of nutrient uptake called the (i) **surge phase**, the (ii) **internally controlled phase**, and the (iii) **externally controlled phase** (Pedersen, 1994).

Active uptake can be described as having three phases:

1. **The Surge Phase**
2. **The Internally Controlled Phase**
3. **The Externally Controlled Phase**

Let's discuss each in detail.

16.4.1 The surge phase



(i) The surge phase

Surge uptake results from a concentration gradient between the alga and the external medium when it is **first exposed** to higher nutrient regimes. This surge in nutrient uptake abruptly ceases several minutes into the experiment as a result of feedback inhibition from pools of inorganic nitrogen and amino acids (as the internal pools fill, the concentration gradient is **decreased** down to a point where the mass influx of nutrient can no longer proceed).

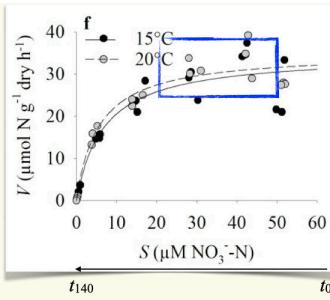
The surge phase is observed at the very beginning of nutrient uptake, at $t = 0$. Imagine taking a seaweed that had not previously been exposed to nutrients and placing it into fresh seawater or a beaker with abundant nutrients. At the start, the environment is nutrient-rich, but the internal pools within the seaweed cells are nutrient-poor.

Immediately after exposure, there is a rapid influx of nutrients—nutrients rush into the cellular pools (like vacuoles) which had been depleted. Once the concentrations equalise, the surge phase ends. This rapid initial movement is the surge phase, driven by a steep concentration gradient.

The surge phase only occurs at the beginning of exposure to high nutrient concentrations. Once the internal pools are filled, the diffusive flux equalises, and the rapid uptake stops.

Note: Be mindful that, on the typical uptake curve, time runs in the opposite direction to substrate concentration; do not confuse the two.

16.4.2 The internally controlled phase



(ii) The internally-controlled (kinetically-) phase

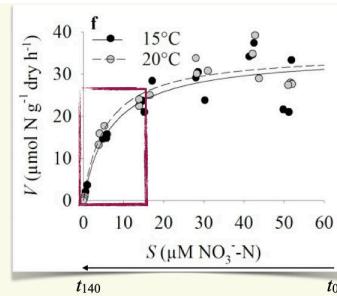
The rate-limiting step is the rate at which N (etc.) is **catalysed to amino acids, macromolecules, etc., and eventually biomass** (this is V_{max}).

Any metabolic process that would require an increased utilisation of amino acids (*etc.*) will maximise the internally-controlled phase.

After the surge phase, once the internal pools are filled, the rate of nutrient conversion—transforming inorganic nutrients already inside the cell into organic compounds (such as amino acids or other macromolecules)—becomes the limiting step. This is the internally controlled phase.

Here, the rate of nutrient uptake is governed by enzyme activity, and this phase sets the plateau seen in the uptake curve (V_{max}). The maximum rate is determined by how quickly the enzymes can process nutrients.

16.4.3 The externally controlled phase



(iii) The externally-controlled phase

A.k.a. the **physically controlled** phase.

The **limit is placed on uptake by the rate by diffusion of N (etc.) across the boundary layer, or...**

...if rate of diffusion is greater than the mass transfer of N (etc.) to the outside of the BL, it is limited by the mass flow of N (etc.).

The limitation of uptake by externally controlled physical factors is called **diffusion transport limitation**, or **mass transport limitation**.

If the plant remains in the closed environment and continues to take up nutrients, eventually the external nutrient concentration will drop. At some point, the uptake rate is determined by the diffusion of nutrients from the environment to the cell—this is the externally controlled phase.

...

The rate of transport of a nutrient from moving water to the algal surface takes place through the boundary layer surrounding the seaweed.

The rate of diffusion is **directly related to the concentration gradient across the boundary layer**, but **inversely proportional to the thickness of the layer**.

Diffusion transport limitation can therefore be alleviated by increasing the concentration of the nutrient in the external medium, or by decreasing the thickness of the boundary layer through increasing the water movement past the thallus.

At very low ambient nutrient concentrations, the uptake rate also becomes low, because diffusion is limited. As nutrient concentrations increase, the concentration gradient increases, and so does

the diffusive flux. In this region, the rate of nutrient uptake is determined by the difference in concentration across the boundary layer surrounding the organism.

Two main factors influence the movement of nutrients across the boundary layer:

1. The concentration gradient between the external environment and the cell.
2. The thickness of the boundary layer, which is influenced by water movement; high water movement results in a thin boundary layer and hence faster diffusion, while low water movement causes a thicker boundary layer which slows diffusion.

All these external physical factors combine to influence the maximum rate of diffusion across the boundary layer.

16.5 Influence of Morphology and Environmental Factors

Certain seaweeds with high surface area:volume ratios—those with flat, membranous, or highly branched forms—are optimised for rapid nutrient uptake. When placed in nutrient-rich medium, these forms respond almost instantly, with rapid increases in biomass, sometimes doubling mass within a day or two, given high V_{max} and sufficient nutrients.

On the other hand, seaweeds with significantly lower surface area:volume ratios respond more slowly. They may exhibit ‘luxury consumption,’ taking up nutrient amounts exceeding immediate growth requirements and storing these for future use when growth conditions permit.

Consequently, the physiological and morphological traits associated with fast uptake—high surface area:volume ratio, high V_{max} , and low K_s —are precisely those that predispose certain species to become nuisance algae under eutrophic conditions.

16.6 Factors Affecting Nitrogen (and Other Nutrient) Uptake

16.7 To summarise, several factors determine nutrient uptake rates:

- **Physical environment:** The concentration of nutrients in the water and the thickness of the boundary layer, affected by water motion.
- **Form of nutrient:** For example, ammonium is absorbed much more rapidly than nitrate. Nitrate must be taken up by active transport, whilst ammonium can diffuse passively into the cell.
- **Nutritional state of the plant:** Nutrient-starved plants will take up nutrients rapidly when re-exposed, while replete plants will not show a significant response.
- **Growth environment:** Higher light intensities and temperatures increase metabolic rates, raising V_{max} and thus enhancing nutrient uptake.
- **Morphology:** As discussed, a higher surface area to volume ratio increases both diffusion and uptake capacities.
- **Uptake mechanism:** Combinations of uptake mechanisms (active, passive, facilitated) determine overall absorption rates.

All of these factors interact to control the rate at which nitrogen, phosphorus, or any other macronutrient can be absorbed from the environment.

17 Passive Uptake

Good morning, everyone. This is our last lecture, so I would just like to wrap up more slides. It's not going to be a very long lecture. What we need to talk about today are the two remaining kinds of uptake mechanisms. We spoke at length about active uptake, which is characterised by the Michaelis-Menten equation, but there are also other kinds of uptake mechanisms, primarily passive uptake and facilitated uptake, and today we're going to quickly talk about both of those.

The reason why we have a different kind of uptake mechanism is because there are various different kinds of nitrogen available in the environment. For some of the more complex molecules like nitrate, active uptake is necessary, but there are more simple molecules also available that make up the total DIN (Dissolved Inorganic Nitrogen) pool, and in this instance we talk about the molecule ammonium or ammonia.

Passive uptake

Passive uptake occurs without the expenditure of metabolic energy.

Usually indicative of the uptake of gasses such as CO₂, O₂ and NH₃ (ammonia), or other **uncharged molecules**.

Such uptake processes are usually not affected by temperature.

The passive uptake mechanism, when we talk about nitrogen uptake, is going to be mostly applicable to the uptake of ammonium from seawater into the seaweed itself. If you want to know a little bit more about nutrient uptake in seaweeds, and it's definitely recommended that you do this, you can read that paper that I wrote about 23 years ago in 2002, that talks about nutrient uptake, and it looks at nutrient uptake of ammonium and nitrate, and the various different rates of external water movement at different temperatures by one particular kind of seaweed. So have a look; it's going to give you nice additional extra information that's necessary, and that might make the difference in your exams, to be able to give me a mark, an answer worth 100% versus an answer worth 80%. So every little bit of additional work that you're going to do, by reading additional papers and so on, is going to count in your favour.

The uptake of ammonium is established in the same way that we do for nitrate uptake. In other words, we apply either multiple flask or perturbation experiments, we establish a depletion curve, and from the depletion curve we derive V versus S , in other words the uptake kinetics graph.

When you plot the uptake kinetics graph for ammonium, you will notice that a straight line best describes the relationship between uptake rate and substrate concentration. Here we see a nice straight line going through all of the points. But in the instance at the bottom, when we look at the uptake of nitrate — also done via initially doing a depletion curve — and when we translate these data into those data, and we try and fit a line, we see that no longer can a linear relationship describe the relationship between V versus S ; it's mostly done via a Michaelis-Menten curve. But in passive uptake, when we try to relate V to S , we're always going to find a linear relationship. That's the primary difference in the uptake kinetics between active uptake and passive uptake. Passive uptake, in the case of ammonium, is always going to give us, when we relate V to S , a linear relationship.

This implies that in passive uptake, no expenditure of metabolic energy is necessary, because all of the uptake process can entirely be described by diffusion, and these usually involve the movement of uncharged molecules. Nitrate is a charged molecule because it has a negative charge; it's got some extra electrons. Ammonia, on the other hand, is a non-charged and uncharged molecule (as is CO_2 , as is oxygen). So uncharged molecules usually diffuse from the external environment into the plant, down the concentration gradient, in other words, from where there's plenty of it in the external culture medium, to where there's less of it inside the plant. So it goes down the concentration gradient. In active uptake, it always goes against the concentration gradient, hence the necessity to use energy to drive that process.

Here, the entire thing relies mostly on diffusion, so therefore also processes external to the cell, external to the thallus, such as the rate of water movement that's going to have an influence on the thickness of that boundary layer, is going to be very important in affecting the rate at which the uptake can take place.

Passive uptake

Note that the nature of the linear relationship describing the passive uptake process **prevents us from calculating the K_s or V_{max} parameters...**

... although it is possible to fit the Michaelis-Menten model to linear data, the parameter estimates obtained is not meaningful in ecological terms because the high concentrations at which the estimated values would fall do not occur in nature.

α : the slope of the linear regression still exists and is **equivalent to α in the non-linear case** (both in its calculation and in the theoretical sense).

V : the slope is also the **same in the linear and MM models**. In MM it is variable over changing S but in a linear model it is constant across S .

Rate-unsaturated uptake has also been demonstrated for some seaweeds for nitrate and urea, but the mechanism here remains unknown.

When we have a linear relationship – here we see the linear relationship – at no point along our increasing range of external concentrations (the substrate concentration) is there any evidence that the rate of uptake is going to slow down. With active uptake, the rate of uptake reaches a maximum, reaches a peak, which is V_{max} , but there is no V_{max} here because it's a straight line. So if you're going to increase this concentration here from 40 to 80 to 120, the line is just going to continue to go up and up and up, which means that the rate of uptake is proportional to the amount of nitrogen present in the external environment only. That's going to set up higher concentrations, it's going to set up a steeper concentration gradient, and when we have a steeper concentration gradient, the rate of diffusion is going to increase.

That is what is meant here: the nature of the linear relationship means that we cannot, like in the case of the Michaelis-Menten relationship, calculate something called K_s or the parameter called V_{max} , because enzymes at no point, internal to the plant, have an influence on the maximum rate of uptake of these things. The K_s relationship, that point where the concentration that is associated with the rate of uptake, which is half of V_{max} , also cannot be calculated, because in order to calculate K_s , we do need a V_{max} .

However, we can calculate a thing called α , and α , in the case of a linear relationship, is simply the slope of that line. The slope of the line is directly related to α – it is α , in fact – so by simply calculating the slope of a linear regression, we can know what α is, and α has the same meaning as in the case of active uptake: it tells us something about the affinity of the plant for a particular nutrient. In other words, the steeper α is, the more rapidly uptake rate is going to respond to a change in nutrient concentration. But with a low α (in other words, a very shallow slope), you need a far larger change in nutrient concentration for it to bring about the same rate of change in uptake rate.

So things with a very steep α , a very steep curve, will show a kind of seaweed that is very able to take up nutrients when the amount of nutrients in the external environment is low. Seaweeds generally with a low α – sorry, a, yes, a less steep slope, in other words, a low α – those are the kinds of seaweeds that if you put them in a low nutrient environment, they are not going to be able to take up nutrients very effectively. Usually, given a particular nutrient concentration, say a nutrient concentration that's very low, and you put a seaweed with a low α right next to a seaweed with a high α in the same low nutrient water, the seaweed with a high α is going to be better able to take nutrients from that seawater, and is better able to sustain its nutritional needs in order for it to continue to grow when the environmental nutrient concentrations are low.

So that's a nice way that you can use the knowledge of the steepness of that slope – in other words, that mathematical relationship that relates the rate of uptake to the amount of nutrients present in the water – that knowledge tells us something about the ecological competitiveness of two different kinds of seaweeds with different alphas in the same nutrient medium. And this also tells us a little bit about which seaweeds are going to become more prone to becoming nuisance seaweeds under eutrophic conditions. So the ones with a high α are going to be the ones that are also going to be able to respond very rapidly to any enhanced amount of nutrients in the environment, and they're going to have a tendency to become a nuisance species when eutrophication happens.

17.1 Uptake Kinetics and the Affinity Coefficient

So, when we have a linear relationship—for example, in passive uptake—at no point along our increasing range of external substrate concentrations is there any evidence that the rate of uptake slows down. In active uptake, the rate of uptake reaches a maximum, which is V_{max} , but in passive uptake, there's no V_{max} , because it's a straight line. If you increase the substrate concentration from 40 to 80 to 120, the line will just continue to go up, which means that the rate of uptake is proportional to the amount of nitrogen present in the external environment.

A higher external concentration sets up a steeper concentration gradient, and when we have a steeper concentration gradient, the rate of diffusion increases. This is why, in a linear relationship, we cannot, as we do in the case of Michaelis-Menten kinetics, calculate the parameters K_s or V_{max} , because enzymes at no point, internal to the plant, influence the maximum rate of uptake in these cases. And the K_s relationship—that is, the substrate concentration at which uptake rate is half of V_{max} —also cannot be calculated, because in order to calculate K_s , we need a V_{max} .

However, we can calculate a parameter called α , and α , in the case of a linear relationship, is simply the slope of that line. The slope of the line is directly equal to α , so by calculating the slope from a linear regression, we can know what α is. Alpha has the same meaning as in the case of active uptake: it tells us about the affinity of the plant for a particular nutrient.

So, the steeper the α , the more rapidly the uptake rate increases with a change in nutrient concentration. By contrast, with a low α , or a very shallow slope, you need a far greater change in nutrient concentration to produce the same change in uptake rate.

Seaweeds with a steep α —that is, a steep curve—are able to take up nutrients efficiently, even when the amount of nutrients in the external environment is low. Seaweeds with a low α —a shallow slope

—will not take up nutrients effectively in low-nutrient environments. So, given a particular low nutrient concentration, if you put a seaweed with a low α next to one with a high α in the same water, the one with the high α will better sustain its nutritional needs and enable continued growth in those conditions.

This is a useful way to use knowledge of the steepness of that slope—in other words, the mathematical relationship that relates uptake rate to nutrient concentration in the water. This knowledge tells us about the ecological competitiveness of two different seaweeds with different alphas, in the same nutrient medium. It also tells us something about seaweeds likely to become nuisance species under eutrophic conditions. Those with a high α can respond rapidly to increased nutrient availability, and may become nuisance species when eutrophication occurs.

17.2 Passive Uptake, Thallus Morphology, and the Monod Equation

Coupling: growth-nutrient

When the addition of nutrient to the culture medium leads to an '**immediate growth response**', growth and nutrient uptake are **coupled**, as is the case with the opportunistic bloom-forming species.

In the case where the seaweed is capable of luxury consumption there will generally be a lag between nutrient uptake (or supply) and growth, and in this case growth is uncoupled from nutrient uptake.

In seaweeds that are predominantly governed by passive uptake, where the main process driving nutrient acquisition is diffusion, we find that their thallus morphology is typically defined by a highly elevated surface area to volume ratio. In such organisms, many—if not all—of the cells are in direct contact with the external environment. As a result, there is very little capacity for the formation of complex internal tissues capable of storing nitrogen in forms such as the various nitrogenous storage compounds, for instance, phycobilins, phycobiliproteins, or other proteins.

The Monod equation

Mathematically, it is equivalent to the Michaelis-Menten equation, but where the Michaelis-Menten relationship describes nutrient uptake rate as a function of substrate concentration, the Monod model **relates growth rate to substrate availability**...

... by assuming that cells are always in equilibrium with their surroundings and that growth is exponential, which it is not always, especially for the more complex algae such as *Gracilaria* and *Ecklonia* that are capable of **luxury consumption**.

The Monod model is **more likely to be applicable to seaweeds with a high SA:V**.

This morphological adaptation means that nitrogen taken up via rapid diffusion—owing to the high surface area to volume ratio—immediately supports growth. The process is such that nutrient uptake directly and instantaneously leads to biomass accumulation. In these situations, we characterise the relationship between the growth response and the preceding nutrient uptake rate as a very tight, direct, linear coupling.

It's important to note, however, that this relationship between nutrient uptake and instantaneous growth is not such that it results in indefinite enhancement of growth across an ever-increasing range of nutrient concentrations. At some point, even though physical diffusion might occur at a fairly high rate, the growth response will become limiting. The reason for this limitation in growth response is that the processes leading to biomass accumulation—namely, cellular growth and multiplication—are very much governed by the maximum rates at which the various enzymes involved in these processes can operate.

So, beyond certain environmental nutrient concentrations, where growth and nutrient uptake are linear, further increases in nutrient concentration will see the growth rate begin to slow down. This is quite similar to what we observe in the Michaelis-Menten uptake response, where the uptake rate slows down at very high nutrient concentrations. This type of relationship, which couples the growth response to environmental nutrient availability, is described by the Monod equation. Functionally, it is quite similar to the Michaelis-Menten equation. The only significant difference is that, whereas the Michaelis-Menten equation shows V_{\max} or V as a function of substrate concentration, S , the Monod equation describes growth rate as a function of S .

These particular seaweeds, with their morphological and physiological adaptations, are especially prone to become problematic during eutrophication events. Because there is such an immediate translation of nutrient enrichment to growth, they can respond very quickly to increased ambient nutrient concentrations and thus proliferate rapidly.

By contrast, in the alternative scenario—where growth is not instantaneously stimulated following nutrient addition—we observe a phenomenon known as luxury consumption. In this mode, nutrients are taken up and stored internally within the seaweed as molecular forms that contribute little to immediate biomass increment. Instead, these stored forms serve principally as a reservoir of nitrogen.

Typically, seaweeds engaging primarily in active uptake fall into this category. The nutrients stored internally can subsequently be re-mobilised during periods when ambient nutrient concentrations decline to sufficiently low levels such that further influx by diffusion or active uptake is heavily constrained. In this way, the seaweed can continue to support growth by drawing upon its internally stored nitrogen reserves, even when external supply is insufficient.

18 Facilitated Uptake Mechanism

Facilitated uptake

Resembles passive transport in that it takes place down an electrochemical gradient.

Carrier proteins or ion channels in the membrane assist in bringing the ions into the cells.

Similar to active transport in that

- (i) it can be saturated and data are described by an Michaelis-Menten-like equation.
- (ii) only specific ions are transported.
- (iii) it is susceptible to competitive and non-competitive inhibition.

The third type of uptake, after active and passive, is facilitated uptake. Facilitated uptake resembles passive uptake in that it moves nutrients down a concentration gradient—the external concentration is greater than inside the cell.

However, unlike passive uptake, which relies entirely on diffusion, facilitated uptake uses a particular membrane protein that spans the cell membrane. This protein has an orientation across the membrane; its active site is external to the plant and specific for a particular molecule—say, for instance, sulphate. It binds to the sulfate outside, then flips around and releases it inside the cell.

In short, a protein collects something from outside, flips its conformation, and releases the molecule inside the cell—this is facilitated uptake. It is similar to passive uptake in being down the concentration gradient, but it is also similar to active uptake in showing a saturation response. That is, there is a maximum external concentration beyond which the transport protein cannot increase the rate

of transport further—there is a V_{\max} . Facilitated uptake is also very specific to particular nutrients, and is susceptible to competitive and non-competitive inhibition. For example, another molecule may compete with the primary substrate (such as sulfate) for the active site, displacing sulphate and preventing its uptake.

That's essentially what facilitated uptake is about, and I will not go any further on that point.

19 Biphasic uptake

Biphasic uptake

A two-phase pattern observed in the rate of nutrient absorption relative to nutrient concentration:

1. First Phase: High-Affinity Uptake

- At low external nutrient concentrations, algae exhibit a high-affinity uptake system. The uptake rate increases sharply as the nutrient concentration rises, reflecting the cell's high demand and efficient uptake mechanisms when nutrients are scarce.
- This phase generally involves active transport mechanisms, where the cells expend energy to absorb nutrients, **driven by specific transport proteins with high affinity for the nutrient**. It's common under nutrient-limited conditions, where the algae need to maximize their nutrient acquisition efficiency.

We begin with the idea of biphasic uptake. The term refers to a two-phase response in the way algae absorb nutrients, depending on how much of the nutrient is available in the environment. What makes this process interesting is that it is not a fixed, rigid pattern—it is an adaptive strategy. By switching between modes of uptake, algae can remain flexible across very different nutrient environments. In nutrient-limited waters, they prioritise efficiency: they use high-affinity systems that allow them to extract and hold onto scarce nitrogen. But when nitrogen is plentiful, the strategy changes. High-affinity uptake, which is energetically costly, becomes unnecessary, and the cells may downregulate these systems to conserve energy, relying instead on lower-affinity mechanisms. In this sense, biphasic uptake reflects a balancing act between efficiency and economy, depending on the ecological context.

If we contrast this with other uptake patterns, the distinctiveness becomes clearer. Many organisms follow a hyperbolic uptake curve, where uptake rises with nutrient concentration but tends to level off—following Michaelis-Menten kinetics. A linear uptake pattern, by contrast, can occur under steady nutrient supply, where uptake increases proportionally with concentration, often constrained by diffusion or simpler transport processes. The biphasic form is more dynamic. It reflects an ecological adjustment: rather than following a single trajectory, algae switch gears when the nutrient

environment changes. This allows them to optimise both metabolic costs and the efficiency of resource acquisition—an economy of effort matched to conditions.

Looking more closely, we can describe the two phases in detail. In the first phase—the high-affinity phase—the system dominates when nutrients are scarce. Uptake increases sharply even at very low concentrations, a response driven by specialised transport proteins with high binding affinity. This is an energetically demanding mode of absorption, but it is also the only way to secure survival when resources are scarce.

Biphasic uptake

A two-phase pattern observed in the rate of nutrient absorption relative to nutrient concentration:

2. Second Phase: Saturation or Low-Affinity Uptake

- When the external nutrient concentration reaches a certain threshold, the uptake rate often plateaus or increases at a slower rate. This lower-affinity phase reflects a shift in the uptake mechanism as nutrient concentrations become more abundant, reducing the need for high-efficiency, high-affinity transport.
- At this stage, the nutrient uptake may **become more passive or diffusional**, requiring less cellular energy. This phase might involve different or additional transport proteins with lower affinity but a higher capacity for nutrient transport, suitable for high-concentration environments.

The **second phase—the low-affinity or saturation phase**—develops as concentrations increase. Uptake rates slow, often plateauing, because the cell no longer needs to operate at maximum efficiency. Here, uptake can become more passive, sometimes resembling diffusion, and the cell expends less energy. Additional transport proteins may be employed, but they typically have lower affinity and higher capacity, fitting environments where nutrients are abundant.

Biphasic uptake

The biphasic uptake response is an **adaptive strategy that allows algae to be flexible across a range of nutrient environments**:

- In nitrogen-limited conditions, the algae prioritise efficient uptake at low concentrations (high-affinity phase).
- When nitrogen is abundant, algae may downregulate high-affinity transporters to conserve energy, relying on lower-affinity mechanisms.

Biphasic uptake

Contrast with Hyperbolic or Linear Uptake Patterns:

- Hyperbolic Uptake: A more typical response, where uptake rates gradually increase with nutrient concentration but tend to saturate (e.g., Michaelis-Menten kinetics).
- Linear Uptake: Occasionally observed under continuous, non-limiting supply, where uptake increases linearly with concentration, often limited by diffusion or simpler transport processes.

The shift from a hyperbolic to a biphasic or linear response often reflects the algae's adjustment to environmental changes, such as nutrient limitation so that it can **optimise its metabolic cost and resource acquisition strategy accordingly**.

Together, these two modes exemplify a flexible ecological design. The biphasic uptake pattern underscores how algae are not passively subject to nutrient conditions but actively restructure their uptake machinery. By doing so, they maintain competitiveness across the shifting gradients of their environments.

20 Factors Modifying Uptake Rates

Nitrogen

Factors affecting uptake rates and kinetics:

- Water movement (boundary layer effects)
- The type of DIN
- Nutritional history
- Light intensity
- Temperature
- SA:V (or functional form; incl. inter-species differences, effects of morphological plasticity, etc.)
- The combination of passive (diffusion) uptake with facilitated uptake —results in a bi-phasic response
- Others...

20.1 Water movement

Water movement

Usually, kinetic studies manipulate S (independent variable). However, **water motion** also affects V (but not V_{max}) via its effect on the boundary layer.

Water motion alters V by acting on α and K_s , the model parameter that describes the phase of uptake that is under physical (external) control (D'Elia & DeBoer, 1978).

The magnitude of V_{max} on the other hand, should be **independent of any external process**, since it represents the maximum rate at which the alga is able to metabolise DIN that is already inside the cells subsequent to physical transport across the BL and cell membrane.

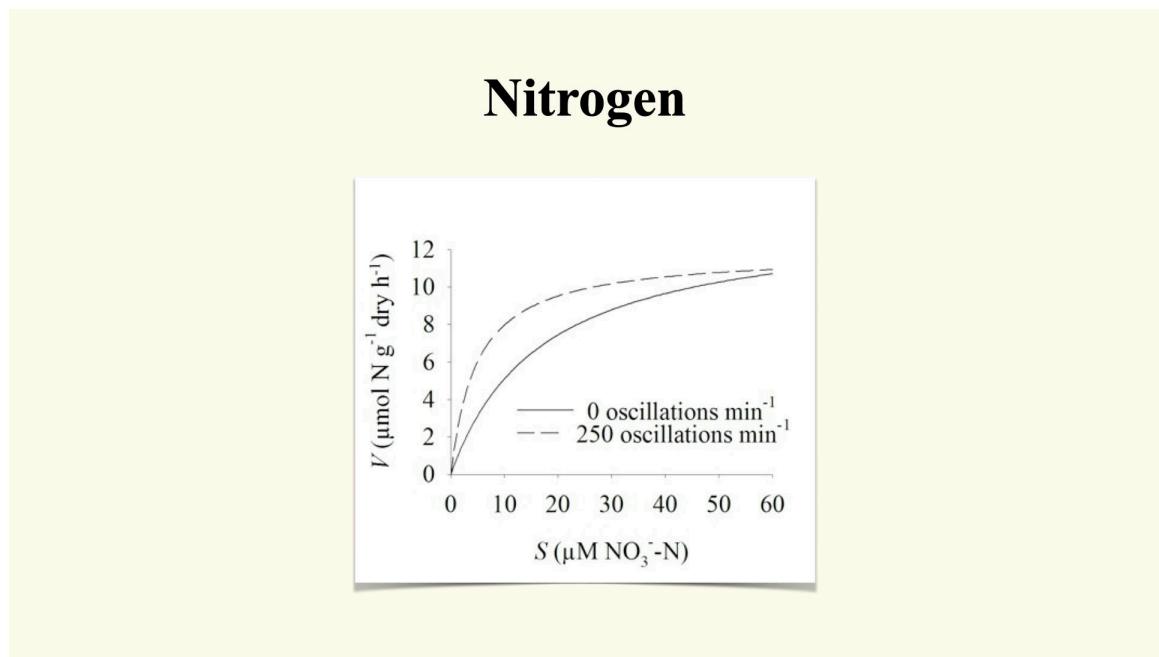
The velocity of nutrient uptake at a given S approaches V_{max} only when the rate of water motion is sufficient to suppress transport limitation; alternatively, an adequately high concentration gradient can overcome transport limitation.

Water movement is an important environmental variable. Consider the ocean: it is highly variable in terms of waviness and the water movement occurring both at the surface and underwater, particularly around seaweeds. Depending on the rate of water movement, there will be a direct effect on the ability of seaweeds to take up nutrients.

Water movement primarily affects the rate parameters— V , the rate of uptake, α , and K_s —but it does not influence V_{\max} , the maximum rate at which nutrient uptake can occur. V_{\max} is always governed by processes operating within the plant itself. In contrast, water motion impacts processes occurring outside of the plant, altering uptake rates via two mechanisms.

The first mechanism is the reduction in the thickness of the boundary layer. A thick boundary layer typically acts as an impediment to nutrient acquisition. With high rates of water movement, the boundary layer becomes thinner, thereby reducing the friction, or resistance, across the interface of the cell wall and the external environment.

The second mechanism is related to the replenishment of localised nutrient depletion. As nutrients are drawn from the immediate vicinity of the seaweed and absorbed into the cells, this can result in a zone of reduced nutrient concentration adjacent to the seaweed. Water movement helps to replace these depleted nutrients—one of the only mechanisms by which this can occur—by enhancing the flow and bringing in new water with a higher concentration of nutrients to occupy the space previously lacking.



Let us consider the effect of water movement on nutrient uptake. To demonstrate this, examine the graph presented here, which contains two traces—one depicted as a dashed line and the other as a solid line.

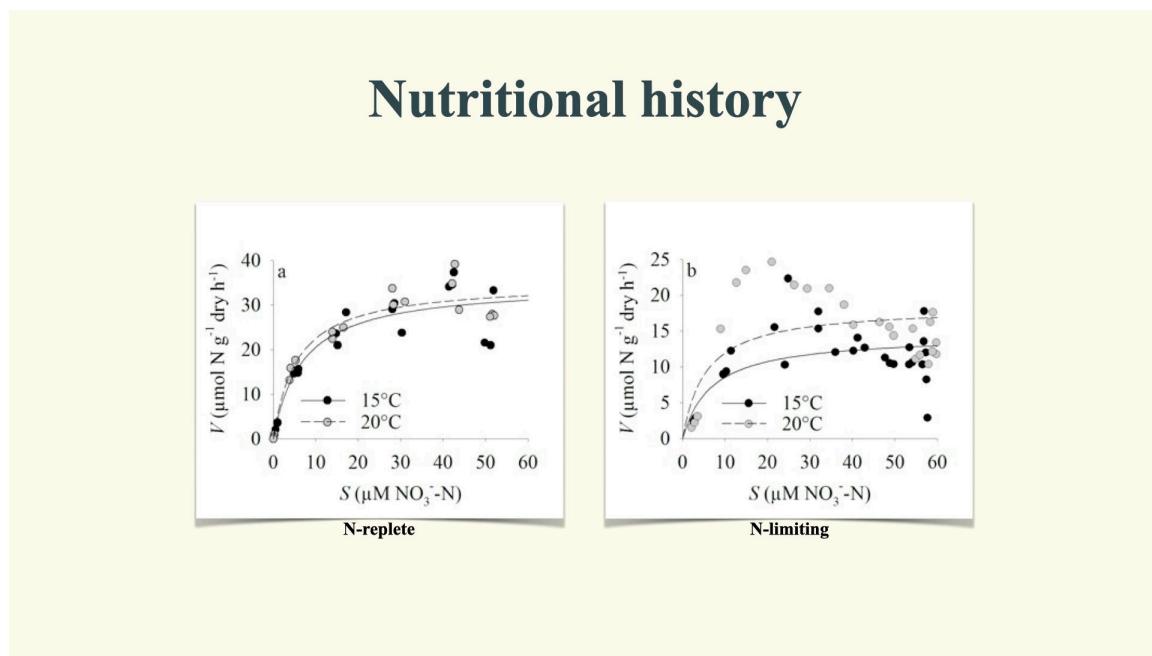
The dashed line represents nutrient uptake in an experiment where glass containers are placed on a shaker table operating at 250 oscillations per minute, intended to emulate an environment with significant water movement. This setting mimics the rate at which water might move around seaweeds in more dynamic aquatic habitats. In contrast, the solid line reflects a condition set to zero oscillations per minute, meaning the water is completely static—absolutely still, with no movement at all around the seaweeds.

Now, if you observe the dashed line, corresponding to 250 oscillations per minute, you'll note that both the rate parameters K_s and α shift. Specifically, K_s decreases to a lower value, and α increases to a higher value. Both of these changes indicate that the rate of diffusion—and therefore nutrient uptake—is significantly enhanced under conditions of elevated water movement.

Conversely, the zero oscillations per minute trace clearly demonstrates a pronounced boundary layer effect. Due to this strong boundary layer, the value of K_s is increased, and the initial slope of the uptake curve, α , is also rather low. Together, these parameters provide clear evidence of the diffusive resistance to nutrient uptake that operates under still water conditions.

20.2 Nutritional history

Typically, when we expose seaweeds – I've already mentioned this in some previous lectures – to a high concentration of nutrients and they continue to be present within that high concentration of nutrients, their rate of uptake is going to slow down because there's no need for them to take up nutrients since all the nutritional requirements have already been met.



But if you take a seaweed from oligotrophic conditions and you remove it from that water and you put it into a new bucket of water with more nutrients in it, it's going to have a very rapid rate of uptake. That's going to increase, in the case of active uptake, it's going to increase the V_{\max} . In the case of passive uptake, it's going to have an influence on the steepness of that line, in other words, α . So the more deprived something is of nutrients, the more it is going to respond in terms of rapidly taking up nutrients when it's placed into an environment where nutrients are quite available.

That's one of the ways in which seaweeds are going to respond to conditions or situations that are going to have a modification on the rate parameters we can estimate from either a linear curve or from a Michaelis-Menten kind of relationship.

Nutritional history

The nutritional history of the seaweed can complicate matters (selection of models, parameter estimation, and interpretation) since it often has a marked effect on the shape of the V vs. S relationship:

- (i) Nitrogen limitation may change the typical hyperbolic response to **a biphasic** (D'Elia & DeBoer, 1978) or linear type (Fujita, 1985). This process is often called rate-unsaturated uptake, and points to an underlying passive uptake (diffusion) mechanism.
- (ii) Nitrogen limitation may increase V_{max} .

In the case of active uptake, this can increase V_{max} . In passive uptake, this influences the steepness of the line— α . The more deprived a seaweed is of nutrients, the greater the response in uptake when exposed to higher nutrient levels.

20.3 Light intensity and photoperiod

Light environment

Light intensity, through its effect on photosynthesis, may influence nutrient uptake rates in active uptake processes:

- (i) it is **responsible for the production of ATP** (energy) that is required for active transport against a concentration gradient.
- (ii) it **provides the C framework** into which the inorganic N is eventually assimilated (proteins, amino acids, DNA, pigments, etc.).
- (iii) **increased growth rate** occurs with increasing light intensity (up to a point).
- (iv) due to **effects on the nitrate reductase enzyme**: the stimulatory effect of light intensity has been demonstrated for nitrate; its absence in ammonium uptake is probably because it is taken up chiefly *via* a passive process.

Another important factor is the light environment. What photosynthesis does is it takes inorganic carbon and puts it inside of the plant and makes it available, converts it into organic carbon. But

in order for organic carbon to be also present as an organic molecule, it needs to be coupled with nitrogen. Remember, organic molecules are always molecules that have in the same molecule, generally, carbon, hydrogen, oxygen, nitrogen, phosphorus; all of those need to be present in order for it to be called an organic molecule. So in order for an organic molecule to be formed inside of the plant, the carbon that's taken up and whose rate of uptake is enhanced under high light conditions, once you put more organic carbon into the plant, it's going to need more nitrogen in order for it to produce organic molecules. So when something produces more carbon, the need for nitrogen uptake increases.

Therefore, typically, when you put a plant into a high light environment or a dim light environment, the ones that are in the high light environment are going to have a higher nutrient uptake rate compared to the ones in dim light. The reasons are explained there. This is true for both linear (passive) and active uptake – more light generally means more rapid photosynthesis.

In the case of active uptake, this is mostly going to affect the rate parameter V_{max} . Coming to linear uptake responses, for passive uptake, the more light there is, the more rapid the rate of photosynthesis is going to be (steeper slope).

Photoperiod

Photoperiod affects nitrate uptake, possibly because **nitrate reductase activity displays a diel periodicity** in terms of activity and synthesis.

Another reason is photoperiod. In other words, whether it is day or night, or how long the light period is compared to the amount of night-time that's available, because the enzyme nitrate reductase – that's the enzyme that's first going to take nitrate once it has been taken up and convert it into ammonium before the ammonium can be translated into amino acids – that activity of the nitrate reductase enzyme is very much coupled to photoperiod. So the more light there is, the more active nitrate reductase is going to be, and the faster the rate of nitrate uptake during light conditions. So with longer light availability, nitrate reductase is more active, and therefore the rate of nitrate uptake is greater.

20.4 Temperature

Temperature

Temperature mostly affects active rather than passive processes.

An increase in temperature increases metabolic rate: active uptake and general cell metabolism has an approx. **Q_{10} of 2** (*i.e.*, a 10°C increase in temperature results in a doubling of the rate.)

For passive (*i.e.*, driven by physical factors) transport, **Q_{10} is much less, typically 1.0 to 1.2**.

Temperature effects are ion-specific and depends on the species.

Another thing is, of course, temperature. I've mentioned this also before. That's because of this thing that's called Q_{10} . This is something that Prof Maritz must have mentioned in one of his lectures when he talks about the rate of metabolism, and typically the rate of metabolism doubles for every 10°C increase in temperature. As the rate of metabolism increases as a function of temperature, it means that more organic carbon can be formed, and as more organic carbon can be formed, it requires more nitrogen to be taken up. Because things grow faster – obviously up until the optimal or maximum temperature – when they grow faster as a result of being exposed to more heat, more nitrogen would also be necessary for it to support the formation of organic molecules. Temperature effects tend to be ion-specific and depend also on the species, so it's going to depend on whether it's taking up nitrate or ammonium, and this effect of temperature to enhance the rate of metabolism is going to very much depend upon the species in question.

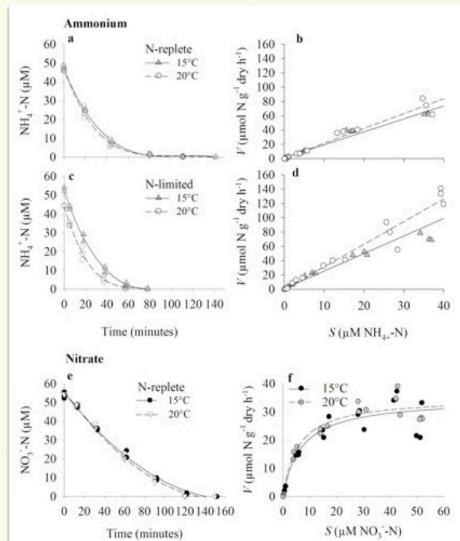
20.5 Nutrient type

Different types of DIN

In some instances the uptake of nutrients (*e.g.* ammonium) does not appear to be saturated even under high experimental concentrations irrespective of past nutritional history and then other models must be sought to describe the relationship (Smit, 2002).

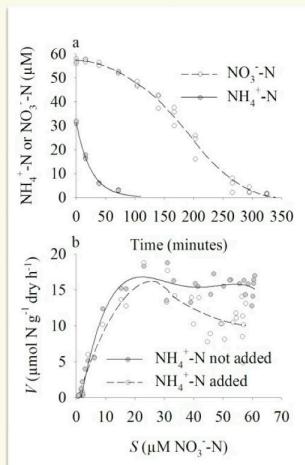
The transport (uptake) rate is directly proportional to the electrochemical-potential gradient (as determined by the difference in concentration between the exterior and interior of the cells).

This is usually the second type of nutrient uptake, *i.e.* passive uptake (unlike the Michaelis-Menten type, which is active).



The type, and the concentration of nutrients also play a role. For instance, the type of nutrient —ammonium versus nitrate—dictates uptake mechanism: ammonium shows a linear, passive mechanism, while nitrate shows a Michaelis-Menten active mechanism.

NH₄⁺ vs. NO₃⁻



Some nutrients interact in their uptake. If ammonium and nitrate are both present in culture medium, seaweeds will preferentially take up ammonium; nitrate uptake does not occur until all ammonium has been depleted.

20.6 Other factors

Other factors affecting uptake

SA:V affects nutrient uptake and explains for much of the variability observed between species (see the functional form model) or even within a species or within one individual (the functional form approach can be extended to different parts of the macroalgal thallus, or to different morphological strains of a species).

Different species have diff. uptake responses, e.g. *Ulva*: linear (passive); *Gracilaria*: Michaelis-Menten (active).

Other factors responsible for influencing uptake rates include **desiccation**, the **type and concentration of the nutrient** (some of this has been discussed), **interactions with other nutrients** (e.g. nitrate uptake is inhibited in the presence of ammonium), **biological interactions** (competition, density), **intrinsic biological factors** (production of hyaline hairs, reproductive state, age of the thallus, changes in morphology, genetic influences).

Other influences include surface area to volume ratio. You need to know, in detail, how the surface area to volume ratio modulates different physiological responses in seaweeds.

There are also various different intrinsic adaptive biological factors, such as the production of hairline hairs. Sometimes some seaweeds, under very oligotrophic conditions, would actually physiologically enhance or change their morphological appearance, and they do this by producing hairline hairs. Hairline hairs are just small little protrusions from the surface of a normally flat algal thallus. As it produces little hairline hairs, it increases the surface area to volume ratio and increases the affinity for nutrients. Therefore, the seaweed becomes more effective at taking up nutrients when the environmental nutrient concentrations are low.

There are also things such as the reproductive state. Some seaweeds, when they become reproductive, would require an enhanced uptake of nutrients in order to sustain gamete and spore production. Sometimes as a seaweed thallus ages into something that's older and grows slower, the rate of nutrient uptake would decrease with time. When it's very young and rapidly growing, the rate of nutrient uptake would have to be high. Sometimes there are seaweeds that change between morphological appearance — they have heteromorphic alternations of generations. One morphological appearance of a seaweed would have a different kind of nutrient uptake response compared to the other morphological appearance in the same species. There can also be, within the same species, different genetic influences that can affect the uptake kinetics of seaweeds.

21 Conclusion

If you need to know more about seaweed nutrient uptake, or nutrient uptake more generally (which can be generalised to plants), do look at the references provided. At the very least, I would like you to read the paper I wrote, as everything I have lectured on around this section is based on those experiments.

And that brings me to the end of this nutrient uptake lecture, and indeed to the end of BDC223 as far as the plant component is concerned.

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