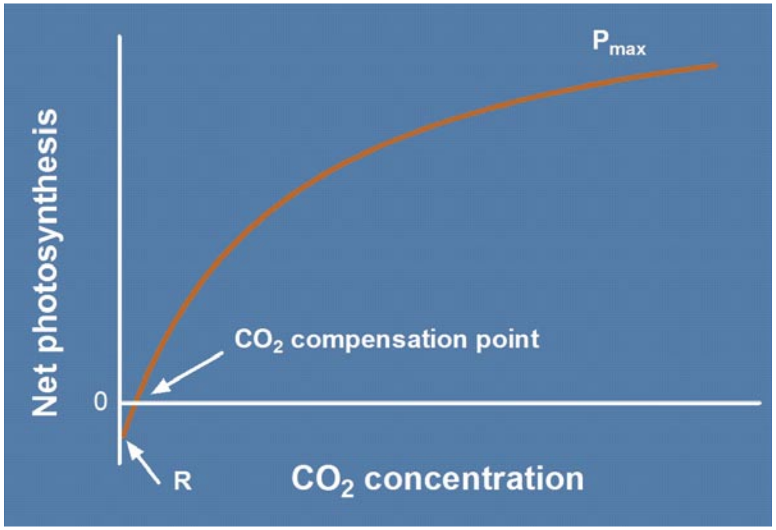
CO_2 in Planted Aquaria

Ole Pedersen[1], Troels Andersen[2] and Claus Christensen[2]

CO_2 is beyond comparison the most important of all plant nutrients. Without sufficient CO_2, plants cannot photosynthesize and convert into energy-rich sugars, starch and all the other carbon-containing molecules that constitute a plant. It may seem odd to look at CO_2 as a plant nutrient. When growing terrestrial plants, we are used to providing light, water and nutrients, but never CO_2. This article explains why we need additional CO_2 in the planted aquarium, how CO_2 is used by aquatic plants, how we can supply CO_2 and how much CO_2 they need.

CO_2—the most important plant nutrient

CO_2 is the most important plant nutrient because of its role in photosynthesis, eventually leading to formation of new leaves and roots. Photosynthesis is a process that is only mastered by photoautotrophs, i.e. organisms that can live with light as the sole source of energy. In photosynthesis, carbon dioxide (CO_2) and water (H_2O) are converted into energy-rich sugar ($C_6H_{12}O_6$) and oxygen (O_2) by means of light energy (see Box 1 on page 26).



Photosynthesis as a function of CO_2. At very low CO_2 concentrations, net photosynthesis is negative. The CO_2 compensation point is defined as the point where net photosynthesis is zero. At higher CO_2 concentrations, photosynthesis gradually saturates because things other than CO_2 start to limit photosynthesis.

It is evident from the photosynthesis equation that only CO_2, water and light energy are needed to fuel the photosynthesis process. Hence it follows that if one of the three main ingredients is missing, photosynthesis will not take place. This seems odd, because we all know of people who are perfectly able to maintain a beautiful planted aquarium without supplemental CO_2. Therefore, CO_2 must be naturally present in the water or else this would be impossible. In biological systems, CO_2 comes from respiration. You could say that respiration is the opposite process of photosynthesis. In respiration, energy is released when sugars are converted into CO_2 and water. Aquatic plants also respire and they do so 24 hours a day. However, while illuminated, most aquatic plants are producing much more organic carbon in photosynthesis than they are burning in respiration. At night, however, there is no photosynthesis because there is no light. Respiration dominates and CO_2 is produced by plants, invertebrates, fish and microorganisms.

The water chemistry of CO_2

CO_2 is easily dissolved in water and the solubility is high. The solubility is almost 1:1 meaning that 1 L of water can contain almost the same amount of CO_2 as 1 L of air when in equilibrium. When CO_2 dissolves in water, it forms an equilibrium between carbonic acid (H_2CO_3), bicarbonate (HCO_3^-) and carbonate (CO_3^{2-}); see Box 1. The balance between carbon dioxide, bicarbonate and carbonate is strongly dependant on pH. At low pH, carbon dioxide dominates and virtually no bicarbonate and carbonate are present, whereas at neutral pH bicarbonate dominates over the two other carbon types. Only at high pH is there a dominance of carbonate. We can take advantage of this fact and manipulate pH to a level that suits us, and thereby obtain a desirable CO_2 concentration in our planted aquaria.

Uptake of carbon dioxide in aquatic plants

All aquatic plants can take up CO_2 directly from the water. When terrestrial plants take up CO_2 from the air, they do so via their stomata. True aquatic plants do not form stomata and their cuticles are also reduced compared to their terrestrial relatives. Consequently, when aquatic plants take up CO_2 from the water, they do so by passive diffusion of CO_2 from the water over the reduced cuticle and into the photosynthetic cells. In aquatic plants, even the epidermis cells contain chloroplast, to reduce the distance from the CO_2 source to the sink.

In water, the uptake of CO_2 is limited by slow diffusion. Diffusion of gasses in water is almost 10,000-fold slower than in air. We can partly compensate for that by raising the CO_2 concentration in the planted aquarium. In most cases, however, we only raise the concentration 100-fold compared to air equilibrium, meaning that our aquatic plants are still limited by slow gaseous diffusion.

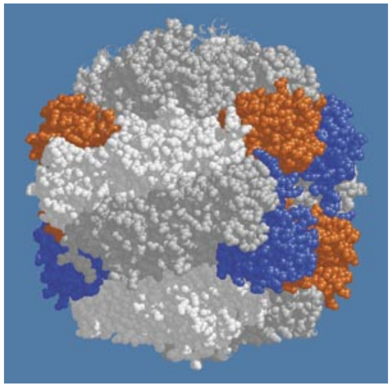
There are alternative photosynthetic pathways and sources of CO_2,

Box 1—

When CO_2 dissolves in water, it forms an equilibrium between carbonic acid (H_2CO_3), bicarbonate (HCO_3^-) and carbonate (CO_3^{2-}) according to **Equation 1:**

$$\mathbf{H_2O + CO_2 \leftrightarrow\ {*}HCO_3 \leftrightarrow\ H^+ + HCO_3^- \leftrightarrow\ H^+ + CO_3^{2-}}$$

It follows from general chemical principles that when the plants use CO_2 in photosynthesis, pH increases because protons are removed from the solution. Protons are removed because the equilibrium tends to flow towards the left when CO_2 is replenished from bicarbonate (HCO_3^-) and carbonate (CO_3^{2-}). The * denotes that this particular process is often catalyzed by carbonic anhydrase, an enzyme that has evolved in plants as well as in animals several times during the evolution of life on earth. On the other hand, at night when respiration dominates, pH will decrease because more protons are formed, when CO_2 is constantly added to the left side of the equilibrium.



A molecule model of the most important enzyme on earth, RuBisCO. RuBisCO is the primary caboxylating enzyme catalyzing the fixation of CO_2 to organic carbon.

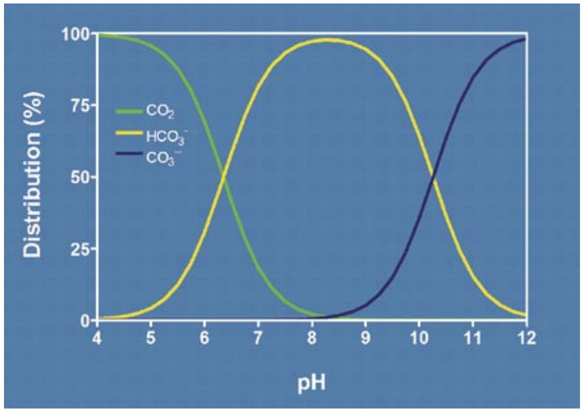
The sum of bicarbonate and carbonate is called the carbonate hardness and is usually measured in degrees (dKH). A better and more correct term is carbonate alkalinity and this is measured in milli equivalents per liter (meq/L). Milli equivalents refer to how many milli equivalents of acid is needed to titrate bicarbonate and carbonate that both act as weak bases.

Once CO_2 has diffused into the cells and further into the chloroplasts, where photosynthesis takes place, CO_2 is converted from inorganic carbon (carbon dioxide, CO_2) to organic carbon (sugar, $C_6H_{12}O_6$) in the photosynthesis according to **Equation 2:**

$$\mathbf{6CO_2\ +\ 12H_2O\ \rightarrow\ 6C_6H_{12}O_6\ +\ 6O_2}$$

Equation 2 is very simplified and in reality, it contains several chemical cycles. To focus on each one of them is beyond the scope of this article. It is important to note, however, that the process must be fueled by light energy and thus, it follows that photosynthesis only takes place during the day.

The first step in photosynthesis is the trapping of CO_2, where the carboxylating enzyme, RuBisCO, catalyzes the first step in a long array of biochemical processes. Like all other enzymes, RuBisCO is made from protein and thus, it has a high content of organic nitrogen. This is the reason why we observe the strong interactions between CO_2, nutrients and light as described elsewhere in the text.



CO_2 as a function of pH. At low pH, most of the inorganic carbon is present as CO_2. At neutral pH, most is present as bicarbonate and at high pH, the equilibrium shifts toward carbonate.

including the use of bicarbonate. These are described in Box 2 page 28.

Interactions with other plant nutrients and light

A high supply of CO_2 can help the plant to conserve other essential nutrients, and if CO_2 is plentiful, aquatic plants can grow even with less light. We discussed this phenomenon in *TAG* Vol. 14 No. 1, Jan. – Mar. 2001, where we used submersed *Riccia fluitans* as a study plant. In brief, our study showed that elevated CO_2 in planted aquaria could maintain the same plant growth but at lower light and nitrogen supply. We concluded that it is often easier to raise the CO_2 level in the tank than to increase the light and thus, we recommend aiming for the higher end of the CO_2 intervals in Box 3 on page 33, particularly if the aquarium is not already well illuminated.

Another aspect of the interactions between CO_2 and other nutrients is that the nutrient levels can be lowered without losing the benefits of CO_2 supplementation. High CO_2 levels in the planted aquarium allow the plants to use less nitrogen for Rubisco, which is the most common enzyme in plants. Rubisco is an enzyme that catalyzes the first step in the Calvin cycle where CO_2 is added to ribulose 1,5 bisphosphate. All enzymes are made from proteins and proteins are very rich in nitrogen. Thus, if less enzyme is needed because the CO_2 concentration is high, the proteins can be used in other processes in the plants leading to formation of new biomass.

Ole Pedersen

CO_2 is the substrate in photosynthesis, whereas O_2 is a waste product. If sufficient CO_2 is present in the water, many plants form oxygen bubbles. Here, *Riccia fluitans* is covered by thousands of bubbles.

CO_2 supplementation in the planted aquarium

If you have an air pump, switch it off! If you have *two* air pumps, switch both of them off! It cannot be overstated that an air pump should never be part of a planted aquarium. The function of an air pump is to supply oxygen (O_2) to fish and invertebrates in aquaria that have no sustainable oxygen production from aquatic plants. In all planted aquaria, there should be more than sufficient oxygen for both fish and invertebrates, even at night when there is no photosynthesis. When plants, fish and invertebrates respire during the night, CO_2 is produced and dissolves readily in the water. This CO_2 can then be used in photosynthesis by the plants once the light is switched on the following day. If an air pump is running, the CO_2 is degassed to the air in the same way carbonic acid is lost from a soda or beer when shaken. Thus, give your air pump a well-deserved retirement!

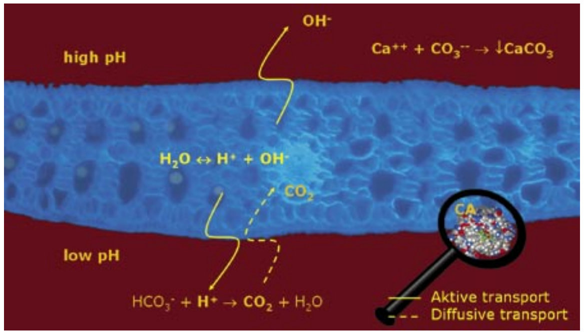
Box 2— Some aquatic plants can use bicarbonate (HCO_3^-) if CO_2 is scarce. In water with a reasonable carbonate alkalinity, bicarbonate (HCO_3^-) is present in large amounts at pH 7–10 (see Box 1 on page 26). On the other hand, gaseous CO_2 is scarce when pH is above 8 regardless of the carbonate alkalinity and thus, aquatic plants that can use bicarbonate as a source of inorganic carbon have a great competitive advantage over strict CO_2 users. The uptake of bicarbonate by aquatic plants is a science on its own, but basically there are two models that explain how it works in most bicarbonate users. One model, first proposed by Prins and Elzenga (1989), deals with plants that have polarized leaves when bicarbonate is utilized. In these plants, protons are pumped out on the lower side of the leaves (abaxial side) resulting in very low pH: down to 4. Here, bicarbonate is converted into CO_2 that subsequently diffuses into the leaves where it is fixed in photosynthesis. Negative loadings in the form of hydroxyl ions are pumped out on the upper leaf surface (adaxial side) where pH rises to above 10. Sometimes the high pH leads to precipitation of calcium carbonate on the leaf surfaces, giving these plants a whitish look. Examples of bicarbonate users are *Elodea canadensis*, *Egeria densa* and most species of pondweeds.


Claus Christensen

During intense photosynthesis, the pH rises on the leaf surfaces, and in some cases it may lead to the precipitation of calcium carbonate. Here, biogenic calcium carbonate has formed on the leaves of *Anubias*.

Other bicarbonate-using plants do not form polarized leaves, for example *Vallisneria* species, and bicarbonate is taken up by the leaves by ion pumps and converted into CO_2 inside the leaves. Regardless of the model in use, bicarbonate uptake is an energy-consuming process and thus, even good bicarbonate users do not produce the necessary enzymes unless needed. In environments with high CO_2, these bicarbonate users cannot use bicarbonate without going though a period of low CO_2 during which the necessary enzymes are produced. One of the most important enzymes in bicarbonate-using plants is the carbonic anhydrase that catalyzes the slow formation of carbonic acid from water and CO_2, or vice versa, which is a critical step when converting from bicarbonate to CO_2.

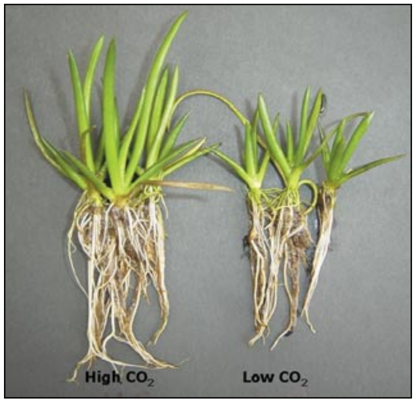
There are a few other tricks that aquatic plants can play to compensate for the slow diffusion of CO_2 in water. One is C4 photosynthesis, which is a common in terrestrial plants. In C4 plants, the oxygen evolving processes are spatially separated from the CO_2 fixing processes. Such plants can photosynthesize at lower CO_2 concentrations because oxygen is kept away from Rubisco. If too much oxygen is present around Rubisco, it turns into an oxygenase, resulting in photorespiration and loss of organic carbon. C4 photosynthesis has only been described for one aquatic plant *(Hydrilla verticillata)* and here it seems to work without the Kranz anatomy that is always characteristic of terrestrial C4 plants. Another strategy that may alleviate the slow diffusion of CO_2 is the dark-fixation of respiratory CO_2 in CAM plants. Here, CO_2 is trapped and stored in malate at night and then subsequently released as CO_2 during the day and fixed in photosynthesis as carbohydrates. Finally, some aquatic plants have specialized in using sediment-derived CO_2 in photosynthesis. Here, CO_2 diffuses from the sediment, where it is present at high concentrations, into the roots and via the aerenchyma further up to the leaves where it is fixed in photosynthesis. Previously, it was thought that this would only be significant in aquatic isoetids (*Lobelia dortmanna*, *Littorella uniflora* and species of *Isoetes*) but recent research by Anders Winkel from the Freshwater Biological Laboratory has shown that sediment-derived CO_2 is also important for photosynthesis in *Vallisneria americana*.



This model shows how bicarbonate uptake works in polarized leaves. Protons are pumped out of the leaves and acidify the abaxial leaf surfaces where bicarbonate is converted into CO_2. The adxial sides are strongly basic often leading to precipitation of calcium carbonate.

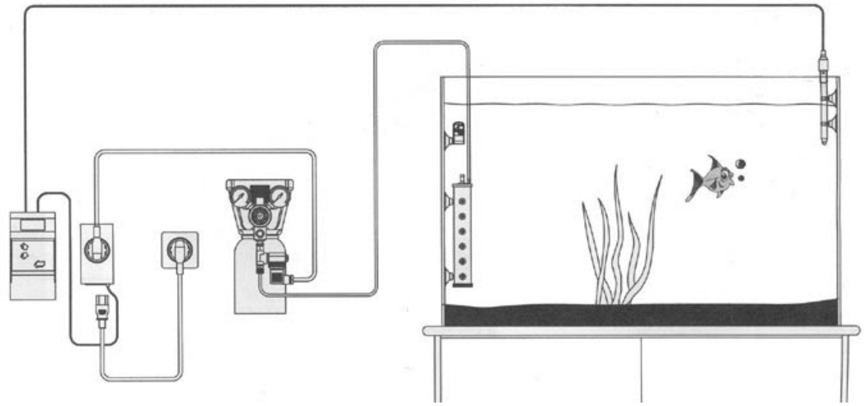
To monitor the CO_2 concentration in the aquarium we recommend using a continuous CO_2 test. It is an ingenious little device that is placed inside the tank, visible from the outside. It works with a chemical color indicator (brom-thymol-blue), which should always be green if the CO_2 level is within the recommended range. In fact the device does not measure CO_2 but pH and thus, it can only be taken as an indication that the CO_2 level is just about right. The dKH vs. pH table in Box 3 shows how much the CO_2 level changes at a given pH as a function of carbonate hardness. The continuous CO_2 test is just an easy way to monitor the CO_2 level (pH) and not a CO_2 supplement in itself.

A yeast reactor is perhaps the cheapest method of CO_2 supplementation. The basic principle of a yeast reactor is based on yeast cells that, in the absence of oxygen, ferment sugar or starch into CO_2. The CO_2 gas is then supplied to the water by means of a bubble stone, mister or CO_2 reactor. The waste product from the fermentation process is alcohol of some sort. (There could be methanol in it too, so don't drink it!) There are numerous designs of well-functioning yeast reactors on the web. Yeast reactors are also manufactured and sold in aquarium shops. A yeast reactor is certainly better than nothing, but it has one major disadvantage and that is that it cannot be easily controlled. Sometimes the yeast cells are happy and ferment a lot of sugar, which leads to huge amounts of CO_2 being dissolved in the water. Other times, the yeast cells are less active and too little CO_2 is supplied to the tank. Some people believe that these CO_2 fluctuations, which lead to similarly high pH fluctuations, have adverse effects on invertebrates, fish and even plants. It may be so in some cases but both lakes and streams in nature may experience huge daily fluctuations in CO_2 and pH. For example, lowland streams in Denmark may have as much as 20 mg per L CO_2 in the morning and only 5 mg/L in the late afternoon. The natural density of aquatic plants is so high that they can extract all this CO_2 during the daytime although it is constantly supplied from the CO_2-rich groundwater. Plants, invertebrates and fish live happily with these dramatic changes in CO_2 over the day. However, some invertebrates and fish may be more sensitive, so one should always check their sensitivity to pH fluctuations in the literature before installing a yeast reactor.



Troels Andersen

Littorella uniflora grown at high and low CO_2. The plants grown at high CO_2 were much larger even though no extra nutrients were added.



A flow diagram showing the different elements in a pH-controlled CO_2 fertilizer system. CO_2 is stored in liquid form in the gas cylinder with a solenoid valve controlled by a pH meter. A pH electrode in the aquarium continuously measures pH and if the plants are using CO_2, pH rises and the pH meter opens the valve. CO_2 flows into the aquarium until pH reaches the set point on the pH meter causing the pH meter to close the valve (diagram by Dupla Aquaristik, Germany).

Various lime tablets that dissolve and release CO_2 when added to the aquarium may also be used as CO_2 fertilizer. We have no personal experience using these products but you can find all kinds of observations on the web reporting either positive or no effects using calcium carbonate tablets in planted aquaria. A few years back, Carbo Plus was marketed. Carbo Plus produces CO_2 electrolytically from solid carbon and it works reasonably well in well-buffered aquaria (8 – 12 dKH carbonate hardness).

Compressed CO_2 is the best method of CO_2 supplementation. When compressed and stored in a tank, CO_2 is liquid and the pressure is approximately 58 bars (approximately 840 psi). CO_2 can be bought in various gas cylinders that are not necessarily custom built for CO_2 fertilization, because CO_2 is also used for soda (for example, SodaStream), welding or as a “propelling system” for tap beer. In principle, all these CO_2 types can be used but in practice, we are limited by the threading of our regulator. In its simplest form, a CO_2 fertilization system using compressed CO_2 consists of a CO_2 cylinder, a pressure reducer with manometer and low pressure regulator connected to a bubble stone, mister or CO_2 reactor.

It is a recurrent discussion whether to switch off the CO_2 supply at night. As explained above, many plants, invertebrates and fish are used to daily fluctuations in CO_2. So, switching the CO_2 off at night is mainly a matter of not wasting CO_2 when the plants cannot use it in photosynthesis.

A somewhat more advanced system may include a solenoid valve that can switch off the supply at night by using a timer. Very advanced systems include a pH electrode and a pH meter that control the solenoid

valve. CO_2 is automatically switched on and off according to a set point. When photosynthesis dominates over respiration, CO_2 is consumed and the pH rises. When the pH rises above the set point, the pH meter opens the solenoid valve and CO_2 is supplied to the water. When sufficient CO_2 is supplied, the pH drops and the pH meter switches off the CO_2 via the solenoid valve. This system conserves CO_2 and keeps the pH very stable. However, the pH electrode must be regularly calibrated to avoid electrode drift that results in CO_2 concentrations far from the desired level.

In planted nano aquaria, sodawater (obviously without lemon and sugar) has been used to boost CO_2. It is not easy to administer and it takes a little experience to get the quantity right. We have seen examples where fish were gasping at the surface following too much sodawater. Also, some plants (for example, *Cryptocoryne sp.*) may be sensitive to dramatic pH changes, so sodawater as a CO_2 fertilizer should be used with caution.

Recently, various organic carbon supplement compounds have been marketed. We have tested two common products on *Hygrophila corymbosa* "Siamensis" (a widespread and popular aquarium plant that only uses CO_2 as a carbon source) and *Egeria densa* (another common aquarium plant able to use bicarbonate) and we found neither positive nor negative effects on the photosynthesis measured as oxygen evolution. Nevertheless, hobbyists have reported positive observations on plant growth when using such organic carbon supplements and a more detailed study is probably required to sort out the pros and cons of these products.

Conclusion

CO_2 is by far the most important plant nutrient. It must be present in reasonable concentrations in planted aquaria. In general, the CO_2 produced biogenically in respiration is insufficient to sustain photosynthesis of aquatic plants that are strict CO_2 users. Thus, CO_2 supplementation is required to grow the more difficult and demanding plants. We recommend 15 – 30 mg/L, although less will show positive effects with most plants. Levels significantly above 30 mg/L can cause adverse effects on invertebrates and fish and thus, the CO_2 must be monitored routinely to maintain a healthy and safe CO_2 level in the aquarium. *Try it now and enjoy how your planted tank flourishes!*

References

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Madsen TV & Sand-Jensen K (1991) Photosynthetic carbon assimilation in aquatic macrophytes. Aquatic Botany 41: 5-40

Prins HBA & Elzenga JTM (1989) Bicarbonate utilization: function and mechanism. Aquatic Botany 34: 59-83

Footnotes

[1] The Freshwater Biological Laboratory, University of Copenhagen, Denmark

[2] Tropica Aquarium Plants, Denmark

Box 3—

Why do aquarium plants need CO_2?

- CO_2 is one of the three ingredients in photosynthesis. Without CO_2, plants cannot photosynthesize and thus, it is the most essential of all plant nutrients.
- In photosynthesis, CO_2 is fixed to energy-rich organic compounds that are used to maintain all metabolisms. Some of the organic compounds are used for growth and thus, plants need CO_2 to form new leaves and roots.

Is CO_2 already present in the aquarium?

- CO_2 is always present in the aquarium but the concentration level is often too low and so growth is strongly limited by CO_2.
- The CO_2 already present in the aquarium derives from the respiration of plants, fish and microorganisms.
- The CO_2 level will be highest just prior to when the light is switched on and lowest toward the end of the light period. This is because the plants use the accumulated CO_2 but it is insufficient to fuel photosynthesis for more than a few hours.

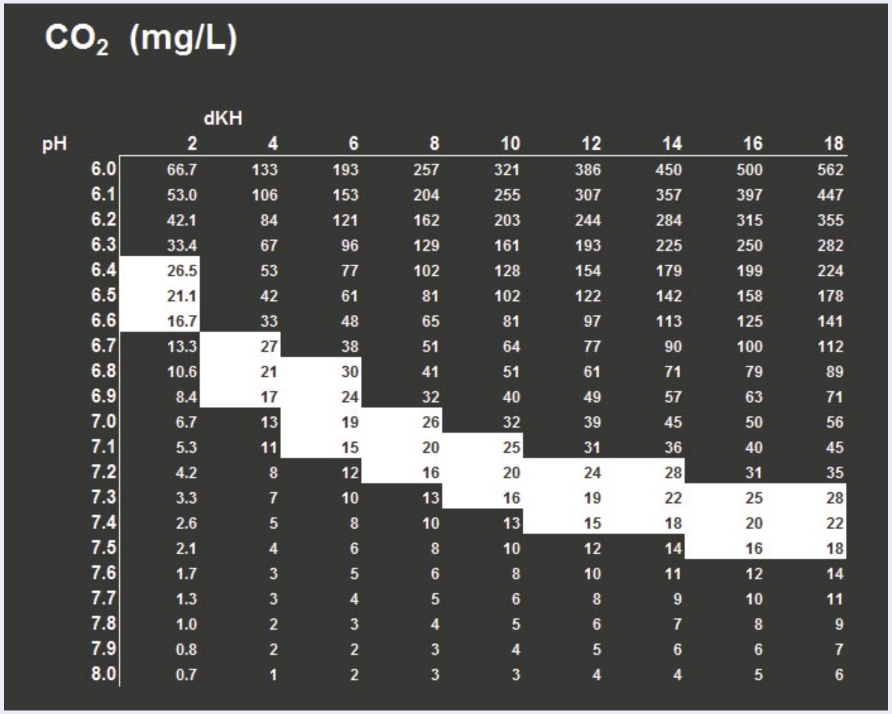
How is CO_2 supplied to the aquarium?

- Yeast reactor
- Lime tablets dissolved with acid
- Electrolytically
- Pressurized CO_2 from a gas cylinder (continuously or via pH controller)

Please note that the relationship between carbonate hardness (dKH) and pH determines the CO_2 concentration in the water. For example, a drop in pH from 8 to 7 in hard water results in significantly more CO_2 than in soft water! Also, make sure that invertebrates and fish are slowly acclimated to water with elevated CO_2.

Which CO_2 level do we recommend?

- Without additional CO_2 supply, the CO_2 level is typically below 5 mg/L and just a tiny increase will result in visible effects on the plants. We recommend aiming for 15–30 mg/L.
- Please note that CO_2 itself also influences pH. The more CO_2 in the water, the lower pH. Aquatic plants easily cope with a pH down to 6, whereas fish and invertebrates may be more sensitive.



CO_2 (mg/L)

	dKH								
pH	2	4	6	8	10	12	14	16	18
6.0	66.7	133	193	257	321	386	450	500	562
6.1	53.0	106	153	204	255	307	357	397	447
6.2	42.1	84	121	162	203	244	284	315	355
6.3	33.4	67	96	129	161	193	225	250	282
6.4	26.5	53	77	102	128	154	179	199	224
6.5	21.1	42	61	81	102	122	142	158	178
6.6	16.7	33	48	65	81	97	113	125	141
6.7	13.3	27	38	51	64	77	90	100	112
6.8	10.6	21	30	41	51	61	71	79	89
6.9	8.4	17	24	32	40	49	57	63	71
7.0	6.7	13	19	26	32	39	45	50	56
7.1	5.3	11	15	20	25	31	36	40	45
7.2	4.2	8	12	16	20	24	28	31	35
7.3	3.3	7	10	13	16	19	22	25	28
7.4	2.6	5	8	10	13	15	18	20	22
7.5	2.1	4	6	8	10	12	14	16	18
7.6	1.7	3	5	6	8	10	11	12	14
7.7	1.3	3	4	5	6	8	9	10	11
7.8	1.0	2	3	4	5	6	7	8	9
7.9	0.8	2	2	3	4	5	6	6	7
8.0	0.7	1	2	3	3	4	4	5	6

This table shows the relationship between carbonate hardness (dKH) and pH. The figures are CO_2 in mg/L. We recommend aiming for 15–30 CO_2 mg/L in planted aquaria.

A Rebuttal and a Response to:

“Light—The Driving Force for the Growth of Aquatic Plants”

CR: The AGA’s esteemed Diana Walstad had some concerns regarding the article by Troels Andersen, Claus Christensen and Ole Pedersen that was published in the last issue of TAG*. She was kind enough to e-mail me her rebuttal. And, well, knowing that* TAG *was all set to publish another article by the same authors, I thought we should give them a chance to rebut the rebuttal. To follow is the text of their worthy scientific debate.*

Diana Walstad writes:

This worthy article was, in my opinion, marred by a couple confusing statements such as the one on page 31 of *TAG* Vol. 20 No. 2: “Also, light is absorbed by colored substances (humic acids absorbing mainly red and infrared light) dissolved in the water and by particles suspended in the water (mostly microscopic algae and detritus absorbing mainly blue light).”

First, water (not humic acids) absorbs red and infrared light. This is a known fact that has been measured by scientists. Wetzel (p. 58) provides the measurements showing that water preferentially absorbs the longer light wavelengths. For example, pure distilled water absorbs 89% of 800 nm infrared light, 28% of 650 nm red light, but only 2% of the 450 nm blue light. Most marine hobbyists intuitively understand that the reason the deep ocean is blue is that the water preferentially absorbs red light. Dissolved humic acids preferentially absorb the shorter wavelengths (both UV and blue light) (Wetzel, pp 762–763).

Second, the authors (p. 31) state that aquatic plants need a dark period each day to “rest.” I’d like to see some evidence (or a less sweeping statement). For example, one study (Polar 1986) shows that duckweed grown for 8 days under an 18 hr light/6 hr dark regimen grew 34% less than that of plants getting continuous light (24 hr light per day). Perhaps the authors were referring to some plants that capture their respiratory CO_2 at night and store it as malate sugar, which they use during the day. However, this specialized strategy is restricted to mostly Isoetid-type plants due to their severely CO_2-limited natural environment [Boston 1989]. It doesn’t apply to most aquarium plants. Furthermore, who knows what an Isoetid plant would do if it were provided with sufficient CO_2 and continuous lighting?

The authors are in the best position to say, “We grow our plants with a 10 – 12 hour rest period, because we found they don’t grow as well with continuous light.” I could accept this statement based on their vast experience, but not the generalized and unsubstantiated statement “plants need a dark period to rest.” Clearly, duckweed do better without a rest!

References

1. Boston HL, Adams MS, and Madsen JD. 1989. Photosynthetic strategies and productivity in aquatic systems. Aquatic Botany 34: 27–57.

2. Polar E and Kucukcezzar R. 1986. Influence of some metal chelators and light regimes on bioaccumulation and toxicity of Cd2+ in duckweed (*Lemna gibba*). Physiol. Plant 66: 87–93.

3. Wetzel RG. 2001. Limnology. Lake and River Ecosystems (Third Ed.). Academic Press (New York), 1006 pages.

Troels Andersen, Claus Christensen and Ole Pedersen respond:

We admit that the way we described light absorption of the various components in water could lead to misinterpretations. Thus, we would like to clarify that:

1) Pure water primarily absorbs infrared and red light.

2) Humic acids primarily absorb in the blue and ultraviolet spectrum.

3) Plankton cells absorb red and blue light following chlorophyll’s spectrum.

4) Most particles scatter the light rather than absorb it.

Regarding whether plants need a period to “rest?” Well, Ms. Walstad provides a convincing example from duckweed that continues forming new biomass even when illuminated 24/7. We could provide examples from microalgae cultures showing that cell division primarily takes place during darkness based on accumulated starch.

However, the formation of new biomass is not our major point. Most processes in plants related to ontogeny, architecture and flowering are controlled by hormones or the phytocrome systems. Phytocromes are molecules sensitive to red light and far-red radiation. Many flowering plants use them to regulate the time of flowering based on the length of day and night and to set circadian rhythms. We admit that the word “rest” is ambiguous and insufficient to explain the advanced processes we had in mind.