

TECHNICAL DATASHEET

The analysis for Chlorophyll 'a' and Phaeophytin

What is Chlorophyll 'a'?

Chlorophyll is the pigment found in plants (and therefore in algae) that is responsible for photosynthesis. Analyses for pigments such as chlorophyll and especially chlorophyll 'a' are widely used to assess the abundance of Algae present in suspension in natural waters. Under favourable conditions chlorophyll 'a' determination is rapid, reproducible and reasonably specific for photosynthetic plant material. Determination of algal biomass can be made difficult using chlorophyll content alone because there can be wide variations in chlorophyll content between plant species and chlorophyll may also be difficult to abstract from the cells of some algal species. Therefore determination of the pigment content of a sample needs to be regarded as a contributing factor to biomass assessment, rather than a definitive technique.

Phaeophytin is one of the breakdown products of Chlorophyll. It remains pigmented, but is not active in photosynthesis.

Phaeophytin therefore can interfere with the chlorophyll analysis because it absorbs light and fluoresces in the same region of the spectrum as chlorophyll 'a' and, if present, may cause errors in chlorophyll 'a' values. In typical water bodies, phaeophytin will contribute between 16% and 60% of the overall absorption. The ratio of chlorophyll 'a' to phaeophytin 'a' serves as a good indicator of the physiological condition of phytoplankton of the sample.

ALS Coventry is able to undertake the analysis of Chlorophyll and Phaeophytin. This analysis for Chlorophyll 'a' is UKAS and DWTS accredited.

The method can be applied to all types of water to determine the concentration of chlorophyll 'a' in a given volume of sample.



The impact of Algae – are they dangerous?

Algal blooms can give rise to water quality and engineering problems such as water discoloration, filter clogging and the generation of taste and odour problems.

Some species produce toxins, which have been associated with wildlife, especially fish mortalities. Thus knowledge of algae abundance, genus quantity and growth potential is of great importance to water treatment personnel responsible for monitoring water quality and treatment efficiency at water treatment works. Blooms in recreational waters can lead to closure which may impact on the recreational use of the associated waters.

A blue-green colour indicates the presence of species of Cyanobacteria which release toxins that may cause serious irritations by skin contact. Deaths of animals such as dogs and sheep have been recorded when they have been allowed to eat the scum. To avoid this hazard, those responsible for water bodies such as the Environment Agency and water companies post warning notices on footpaths around rivers and reservoirs where recreational activities are permitted. These notices warn of the need to avoid direct contact with the water. At such times various water contact activities may be prohibited such as diving, sailing, kayaking and canoeing. Poisoning due to ingestion of algal toxins is a very rare event and

Do Algal blooms affect the safety of drinking water?

those that are recorded occurred in other parts of the world such as South America, Australia and the Far East. These harmful events were linked to extreme climatic conditions or unsuitable private water supplies without adequate management or treatment.

Cyanobacteria (Blue green Algae) can produce toxins.

Studies in the UK demonstrated that conventional water treatment is effective at removing algae and their associated toxins.

By law, water companies must identify all water sources at risk from algal blooms. These sources must have adequate treatment in place to safeguard the quality of drinking water in the event of an algal bloom. Water companies monitor river and reservoir water for the presence of algae by counting cells under a microscope and measuring the amount of chlorophyll in the water. These tests give early warning of bloom conditions. Water companies have the capability to test for common toxins such as Microcystin – LR and the World Health Organisation has set a provisional health related guideline value of 1 µg/l. Toxins are not routinely tested for in drinking water but testing can be carried out to check that water

treatment is effective under bloom conditions. ALS Environmental is able to undertake Microcystin analysis at ALS Wakefield.



Method

The ALS method for the detection of Chlorophyll 'a' and Phaeophytin is suitable for the analysis of treated and surface waters.

Plant material such as Algae is obtained from a water sample by an initial filtration step. Solvent extraction of the pigments is achieved using methanol. (Methanol extraction is particularly useful as it is superior to acetone as an extractant especially when hot). Finally, the chlorophyll 'a' concentration is determined by spectrophotometric evaluation of the extract by carrying out absorbance measurements at two wavelengths: 665nm offers the maximum absorption of chlorophyll 'a', while 750nm compensates for 'background turbidity'. Acidification of the sample will denature and convert chlorophyll to Chlorophyllide and the re-read will relate to the original phaeophytin levels.

Both Chlorophyll and Phaeophytin are pigmented and will contribute to the initial spectrometric result.

Rapid Acidification converts Chlorophyll to Chlorophyllide. This is unpigmented. Initial spectrophotometry will evaluate both pigments, while only phaeophytin will remain after acidification.