



**To test the presence/absence of microalgae in water samples
Used in oil, gas and petrochemical industries, air industries,
food industries, water and waste water, other industries
with special formulations to check different samples
including drinking water, waste water samples and food
samples.**

Microalgae have simple and undifferentiated cell structures. They grow slowly and exist in nature as attached to surfaces or free (planktonic). In general, the probability of the presence of planktonic algae in water samples is higher unless the algae attached to the surfaces are separated due to the change of water flow or any other physical factor and enter the water samples. Therefore, depending on the type of algae under study, one should be careful in sampling methods. By continuously testing water and waste water, for example, monthly, using this technique, checked the level of possible microalgae aggression.

The culture medium used contains essential nutrients for the growth of algae such as nitrogen, phosphorus, potassium, sulfur, etc. Microalgae usually grow slower than heterotrophic bacteria and require light for photosynthesis. Most of them can use low light levels.

Due to the slow growth of microalgae, it can be difficult to detect the beginning of growth. Therefore, microscopic examinations can determine the first signs of growth earlier. Many microalgae are seen in green color at the beginning of their growth due to the production of chlorophyll. As the growth continues, other pigments such as xanthophylls may dominant over chlorophyll and the color of the growth will change. Note that this color change can include green to yellow, orange or brown colors. The growth of microalgae also occurs regionally or throughout the culture medium.

The MicrobCheck™ Algae test kit is designed as a 50 ml falcon containing agar and broth culture medium.

Manufacturer's Recommendation

Avoid contact with the inner wall of the falcon. Perform the test under sterile conditions.

After opening the falcon, place the door upside down, with the bottom facing the ground, on a clean surface.

Before using the kit, shake the water sample well.

Test Method

Preparation

Collect at least 25 ml of sample.

Pour 19 ml of the sample into the Falcon and close it.

Write down the date and name of the sample on the falcon.



Note that if it is necessary to check the population of microalgae in soil or slurry, it is necessary to make changes in the method of using and inoculating the test kit. Because there is a possibility of the growth of heterotrophic microorganisms due to the high amount of nutrients in the soil. Take 1 gram of soil or slurry and dissolve it in 9 ml of sterile Ringer's solution. Mix well to form a homogeneous solution. Take 1 ml from the middle of this suspension and add it to 14 ml of sterile Ringer's solution. Transfer this total of 15 ml to the falcon test kit and heat it. In this method, it is necessary to multiply the calculated live population by 2 to get the possible population in the original soil sample.

Incubation

incubate the inoculated falcon horizontally under the light of a 40-watt lamp at a distance of 60 cm and at room temperature (21-25 °C).

Check the samples at least three times a week for 25 days.

Check the samples for growth and color change. Note the date of the first observed reaction.

Note that to avoid overheating the test kit because a temperature above 30 °C inhibits the activity of microalgae. Also, under no circumstances should the falcons be shaken during the test period.

Note that microalgae grow slowly, that's why it is difficult to detect the beginning of growth. Examining a small amount of the medium under the microscope can help to better determine the growth. Some algae are seen in green color at the beginning of their growth. This problem is caused by the production of chlorophyll pigment that they produce for photosynthesis. As the growth continues, other pigments such as xanthophylls dominate over chlorophyll, which is associated with a change in color to yellow, orange, or brown.

Presence / Absence

A positive test is associated with coloured cloudy structures in the broth medium or coloured or non-coloured sections on the agar medium. Note that the growth of different algae is different and not all of them have the same appearance results.

In the Picture, the growth of *chlorella* algae can be seen on the agar medium as well as broth.



Microalgae Growth Patterns

GG - Grass Green growth: If this reaction is observed, the growth of green clots may also be observed in the broth medium.

FG - Fuzzy Green Patches: This reaction is often observed on the broth.

OB - Red Orange Brown Patches: This pattern will change color with further growth.

YB - Yellow Beige Patches: Pale yellow to beige patches that grow on agar and are difficult to observe and identify.

GF - Green Flocculent: Flocculent sediments that are observed in the broth medium and also on the wall of the falcon.



These structures are dense and may be green in color.

DG - Dark Green to Black Patches: Dark green, green-blue or black patches are seen on the surface of the broth medium. Usually, DG reaction is followed by GG, FG or GF.

Due to the fact that the sample can be contaminated with different microalgae, a chain of these reactions occurs one after the other, each of which indicates a dominant population of algae.

GG – DG	The presence of <i>cyanobacteria</i> with the possibility of a dominant population of <i>Nostoc</i>
FG – DG	Green algae with the presence of <i>cyanobacteria</i>
FG – OB	Green algae
YB – OB	Dominant population of diatoms and desmids
GG – GF	Green algae without pigment production
GG – GF – DG	Dominant populations of green algae and <i>cyanobacteria</i>

Estimation of Population and Aggression Level

If the test result is positive, you can estimate the algae population and their aggression level using the table. A faster reaction occurs when the algae population is high.

Aggression Level	Time Lag (day)	Population (cfu/ml)
Extremely High Aggressive	4	100,000
Very High Aggressive	8	20,000
Very High Aggressive	12	7,500
High Aggressive	16	600
High Aggressive	20	100
Moderately Aggressive	24	20
Normal Background	25	Low

Quality Control of the MicrobCheck™ Algae Test Kit

To confirm the quality and performance of the MicrobCheck™ Algae test kit, the specified algae can be cultured and the results checked. Keep the kit at room temperature and under proper light and observe the growth for 25 days.

Organism (ATCC)	Pattern
<i>Chlorella sp.</i>	Growth on agar and broth with green color
<i>Nannochloropsis sp.</i>	Growth on agar and broth with green color
<i>Chlamydomonas sp.</i>	Clot growth in broth and agar medium - white to green color

Best Time to Use

The expiration date of the kits is 6 months and it is necessary to store them in the refrigerator (4-8°C). It is recommended to avoid frequent temperature changes and to avoid freezing.

Disposal

Test kits are completely contaminated after use and algae growth. As a result, they need to be autoclaved or burn in a furnace. If this is not possible, open the falcons under the laboratory hood and fill it with bleach liquid with a concentration of 5 to 10%. Let it sit overnight and then throw it away.

