

## GENERAL PRINCIPLES FOR THE PREPARATION OF NUTRIENT MEDIA REQUIRED FOR THE STUDY OF SOIL MICROORGANISMS

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### ANNOTATION

This article discusses issues related to soil microorganisms, their nutrient media necessary for their isolation and reproduction, in addition to the fact that these nutrient media are a source of nutrients, they are also chemical and physical conditions necessary for the life of microorganisms.

**Keywords:** nutrient medium, microorganisms, nutrient medium, concentration, sterility, soil particles, substrate, soil.

The nutrient medium is necessary for the isolation and reproduction of microorganisms from the soil. In addition to being a source of nutrients, these nutrient media are the chemical and physical conditions necessary for the life of microorganisms. Nutrient media must contain various components, as well as have a precisely measured concentration. The ratio of individual components must be at the optimal concentration necessary for a bacterial cell. If a particular compound is considered as a source. Carbon and energy at the same time, then its amount in the substrate should cover the amount spent on energy consumption and biosynthesis.

To date, there are several groups of microbiological nutrient media. According to their composition, they are divided into natural, semi-synthetic and synthetic types. Depending on the purpose of the study, it is divided into selective and differential-diagnostic nutrient media. Depending on the consistency, it is divided into liquid and solid nutrients.

Large conical flasks are used to prepare the nutrient medium. When incubating cultures of microorganisms, large conical flasks, test tubes or special horizontal vials are used. The heated agarized nutrient medium is placed in test tubes using a measuring funnel. To do this, a funnel of 200 ml is placed on a laboratory tripod, and a 10-centimeter rubber tube is placed in its lower part and a glass tip is put on its end.

The use of clean flasks and test tubes is the most important requirement. Glass containers, washed with various means or chemical mixtures, are rinsed first with hydrogen water, and then with distilled water.

- It is necessary to use pure chemicals. If possible, chemically pure chemicals should be used, especially if the nutrient medium is used to preserve and identify microorganisms.

- When preparing the food medium, it is necessary to add its components in the exact amount. Large quantities of substances should be weighed on technical scales, and small amounts of substances on analytical balances. If it is necessary to measure a small amount of a substance, measure its tenth or hundredth part and dissolve in the required amount of distilled water. The required amount of the substance is added to the nutrient medium using a pipette.

- Individual components are added after the first is completely dissolved in the sequence specified in the recipe of the nutrient medium. Agar is melted in a water bath or steam bath, it is not allowed to melt it on an open fire.
- After preparation, the nutrient medium is checked and, if necessary, the chemical composition of the prepared solution is adjusted.
- During sterilization, the autoclave temperature and sterilization time should be carried out in accordance with the instructions;
- After sterilization, it is necessary to check the pH of the nutrient medium.
- to adjust the pH of the nutrient medium, a 0.1 M solution of caustic soda or acetic acid is usually used;
- It is required to check the sterility of the prepared nutrient medium. After sterilization, the prepared nutrient medium is placed in a thermostat at 28 ° C for 2-3 days.
- Food media should be stored in a cool and dry place.
- Nutrient media, poured into test tubes, can not be stored for more than 1-2 weeks.

Cultivation of microorganisms should always be carried out on a freshly prepared nutrient medium. In recent years, ready-made nutrient media have gone on sale, which greatly simplifies the work and contributes to the standardization of microbiological research methods. Soil is the habitat of many organisms. The organisms that live in the soil are called pedobiontami. The smallest of them are bacteria, algae, fungi and unicellular organisms that live in soil water. Up to  $10^{14}$  organisms can live in one  $m^3$  of space. Invertebrate animals, such as spiders, beetles and earthworms, live in soil air. They feed on plant debris, mycelium and other organisms. Vertebrates also live in the soil. these include rodents - voles, mice and others. They are very well adapted to life in completely dark soil, so they have excellent hearing and almost no vision.

The heterogeneity of the soil causes it to act as a different environment for organisms of different sizes. A system of soil microreservoirs for small soil animals (protozoa, nematodes, etc.), united under the name of nanofauna.

To slightly larger air-breathing animals, the soil looks like a system of small caves. These animals are called microfauna. The size of the representatives of the soil microfauna is from ten to 2-3 mm. Large soil animals with body sizes from 2 to 20 mm are called representatives of the meso fauna. These are insect larvae, millipedes, enchytraeids, earthworms, etc. For them, the soil is a dense environment that provides significant mechanical resistance to movement. These relatively large forms move through the soil or push soil particles apart, expanding natural wells or making new passages.

Soil particle method. If the studied group of microorganisms is present in the soil in small quantities, they can be isolated by placing soil particles on the surface of an agarized nutrient medium without obtaining an enriched culture. Usually take 0.5-0.10 g of soil, mix with water to the state of paste and place parallel to the surface of the agar. Of course, it is necessary to plant in several parallel cups. The stencil on the back of the cup makes it easy and even to apply soil particles. Of course, planting should be done in several parallel cups.

After the growth of microorganisms, the percentage of soil particles in which a certain group of microorganisms grew is calculated, if it is necessary to study these microorganisms in depth or obtain their pure culture, microorganisms can be taken from the surrounding soil particles.

This method is used for the isolation and quantification of azobacteria, nitrifications, cellulose-destroying microorganisms, fungi, yeast belonging to the genus *Lipomyces* from the soil, as well as for the detection of bacteria from poor soils, such as sand.

When nitrogen bacterium is detected, soil particles are planted on the surface of Ashby or other nutrient media where microorganisms grow well. To do this, 100 mg of soil is liquefied to a paste-like state and with the help of a bacteriological loop is placed on the surface of two petri dishes, and it is considered desirable that each cup has 50 particles. The amount of lipomycetes from the soil was also determined by seeding on ashby medium containing sucrose. After planting, the cups are placed in a damp chamber at 26-28 ° C. Nitrogen bacterium counting is carried out after 6-10 days, lipomycetes after 15-20 days.

Silicic acid gel or unenriched agar is used in the study of microorganisms that decompose cellulose. The next nutrient medium consists of distilled water with 1.5-20% agar, for which filter paper cut into circles is placed on the surface of the agar, thoroughly washed and soaked in the environment of Vinogradsky or Hutchinson. 25 pieces of soil are laid on it. The cups are placed in a damp chamber at a temperature of 25-30 ° C and the growth of microorganisms is observed. They count the number of soil particles in which microorganisms have developed, and determine the nature of the microorganisms grown in them.

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