Opinion

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The Challenge in Identification of Bacteria and Fungi in Environmental Microbiology

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Microbes can thrive in a variety of environments, extreme environments as well as normal environments in soil and water where most of the other species are found. The ability to adapt in various habitats confirms why bacteria, especially prokaryotes, are the most abundant organisms on earth. Microbial ecosystems consist of terrestrial and aquatic, within the human body and in man-made structure. According to a study in 1998, the number of bacteria on earth is estimated at 4-6 x 10^{30}. About 3,5 x 10^{30} prokaryotes live in the deep biosphere in the ocean and 0,25-2,5 x 10^{30} in the deep biosphere on the terrestrial. Lower numbers were found in terrestrial habitats, at 2,6 x 10^{29} cells, and in aquatic habitats, 1,2 x 10^{29} cells. Much smaller proportions are found in the bodies of animals (including humans), in leaves and other plant parts and in the air [1-3].

Related to this, the science of environmental microbiology has developed, namely the branch of biology that studies the interactions between microorganisms, the earth and atmosphere. Then developed into air, aquatic and soil microbiology which plays role in agriculture, industry, fisheries and health. The development of this science has brought progress in discovery of new microbes but of the many existing microbes only about 1% have been identified. Air microbiology deals with microorganisms present in the air. The air is an intermediary medium for several microorganisms that attack humans, animals and plants. The small size of microorganism is easy to move so that it is possible to be inhaled, ingested or due to direct contact, clinically it can cause infection. So far, researchers have revealed that the most commonly found microbes were *Bacillus, Aspergillus* and *Rhizopus*. Unfortunately, the dominant microbe was only identified up to genus.

WHO has set a maximum microbial limit in hospital rooms of 150 CFU/m^3. Several countries have their own standards, such as Brazil, which requires a maximum amount of fungal contamination in rooms where critical patients are treated, which is 750 CFU/m^3. Meanwhile, in Portugal and Canada, the maximum amount of fungal contamination in critical rooms in hospital is set at 500 CFU/m^3 and 150 CFU/m^3, respectively. In Indonesia the microbial limit in the intensive care unit is 200 CFU/m^3 and have to cautious taken carefully especially if pathogens are found and potential to cause disease. A few researchers revealed that the dominant airborne microbes found were *Bacillus* and *Aspergillus*. AS with air microbes, water microbes also important things to study. The main types of water based on location consists of groundwater, originating from deep wells and underground springs and surface water, like rivers, lake and shallow wells. Sources of water microorganisms are air, soil, garbage, mud and plants: organic matter; carcasses of animal and human excrement. Water in nature can certainly be a carrier medium for pathogenic microorganisms which is harmful to health.

Regulation regarding water quality requirements have been established by WHO. For Potable water, WHO recommends a threshold of 0 CFU (Colony Forming Unit)/100 mL of *E. coli*, however, the WHO understand that achieving 0 CFU/100 mL *E. coli* may be challenging in many developing countries and rural areas. Research in Indonesia on the identification of bacteria in refill drinking water depots revealed that Bacillus was the dominant bacteria found. Regarding soil, it consists of organic, inorganic, water and air, all of which are mixed together so that it is difficult to separate one from the other. Microorganisms found in the soil are very diverse, one gram of soil can contain 1010 bacteria cells with an estimated diversity of around 4.103-5.104 species. Microbes found in soil include *Bacillus, Penicillium, Trichoderma, Aspergillus, Mucor, Agrobacterium* and *Rhizobium*. These microbes are difficult to identify with existing automated methods [4-7].

Unidentified microbial diversity is a challenge in itself to obtain an appropriate identification technique. Currently, conventional, semi-automatic and automatic identification methods have been developed. Limitations of conventional methods are not specific to species, require a long time, cannot be used for microorganisms that have not been identified, sometimes faced with microorganisms that exhibit biochemical characteristics that do not match the pattern of every known genus and species, sometimes faced with microorganisms which shows biochemical characteristics that do not match the pattern of each known genus and species. Physical and biochemical characteristics are not static and can change due to stress and evolution, so this also affects the results of identification. The development of semi-automatic and automatic methods has helped a lot in the identification of microorganisms, but several studies revealed the accuracy of semi-automatic machines up to 70 % for the genus level while the species level was only 50%. Several studies revealed that automatic machines can increase the accuracy up to 97,8% for genus and 70,97% for species. It is challenging to find alternative diagnostic techniques fast, precise and economic both in terms of labor and cost, able to detect compounds resulting from metabolism such as carbohydrate, fatty acid, amino acids, and other organic compounds from animal or plant tissues, can be used for the purpose of microbial identification, epidemiological studies, pathogen detection water and environmental samples [8-11].

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