USE OF ACTIVE MICROORGANISM IN IMPROVING POND WATER QUALITY: CASE STUDY OF BEYTEPE POND

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Abstract
Water resources have been seriously polluted in terms of quality in the last hundred years, especially due to anthropogenic effects. The quality of the water in the storage structures (dam, lake, pond, etc.) has started to deteriorate due to the deterioration in the drainage basin, especially the insufficient feeding. In recent years, researches on the protection and improvement of the quality of water in storage structures have begun to increase. In this study, it was aimed to improve the water quality of Beytepe Pond located in the campus of the Turkish National Botanic Garden Directorate (TNBG) by using Active Microorganism (EM) in laboratory conditions. In the study, Baykal EM1, active microorganism, was used. For improvement water quality was used aerobic (A) and anaerobic (AN) systems in containers with a volume of 10 liters. EM was administered in 3 doses as 5, 10 and 20 ml L⁻¹. pH, conductivity (EC), Dissolved Oxygen (DO), Chemical Oxygen Demand (COD) and chlorophyll-a values were measured in the pond water. Beytepe Pond water is 3rd class according to the US salinity laboratory salinity classification (USSL). COD and chlorophyll-a values exceed eutrophication limit values. At the beginning of the study, the raw water COD value was measured as 263 mg L⁻¹. It was determined that 5 ml L⁻¹ EM application was reduced up to 2 mg L⁻¹ in anaerobic system application. The same application provided the best improvement in chlorophyll-a values. As a result of the study, it was observed that the EM application provided an improvement in the quality of the Beytepe Pond water.

Keywords: water quality, landscape irrigation, pond water

1. INTRODUCTION
The continuation of the human race is closely related to the living standards and environmental quality of our planet. Rapid population growth, developments of the industry and technology are putting pressures on the natural resources such as soil and water. Depletion of the amount of useable water and deterioration in water quality are appeared as a result of wastewater discharges from domestic and industrial sources into receiving environments. In order to protect the water resources, the current water quality must first be determined and the water resources should be used correctly (Azaza et al. 2010). In addition, the water resources should be monitored periodically to decide the quality of the usage purpose. In the last decades, problems with water quality have often

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been neglected because of the quality water resources are abundant and easily available (Shamsad and Islam, 2005). However, as environmental problems are increasing in many regions of the world, water quality problems have become more important than the amount of water (Balachandar et al., 2010).

Various chemical and biological applications are available to improve water quality level. In recent years, there has been a technology that uses multicultural mixtures of anaerobic and aerobic beneficial microorganisms. Due to its environmentally friendly nature, effective microorganism (EM) technology is currently gaining popularity. This technology uses naturally occurring microorganisms that can purify and revitalize nature (Zakary et al., 2010). Effective Microorganisms are mixed cultures of beneficial, naturally occurring organisms that can be applied as inoculators to increase the microbial diversity of the soil ecosystem. They consist mainly of photosynthesis bacteria, lactic acid bacteria, yeasts, actinomycetes and fermenting fungi. EM improves the structure of the soil, increases its productivity and promotes biodiversity. It neutralizes pathogens. It increases the intake of nutrients by fixing nitrogen in the soil. It accelerates decomposition and composting by increasing beneficial minerals in organic compounds (Himangini et al., 2019).

Effective Microorganisms (EM) technology was developed in the 1970s at Ryukyus University, Okinawa, Japan (Sangakkara, 2002). Studies have shown that EM can be used in a number of applications including agriculture, livestock, horticulture and landscaping, composting, bioremediation, cleaning septic tanks, algae control (EM Technology, 1998). Effective Microorganisms have been used for solutions to environmental problems. These solutions, which include naturally occurring microorganisms, are capable of reducing the biological toxicity of wastewater while removing bad odours. Therefore, EM has been used in many cases in Japan, especially in Okinawa, to clean wastewater from urban services (Anonymous, 2022a).

In a study investigating the effects of EM on pH and biological properties, it is seen that applying EM to wastewater reduces pH from 7.63 to normal levels (pH=7). Initially, BOD and COD values were 104.5-34.10 mgL⁻¹, respectively analysed as 6.9-27 mgL⁻¹ as a result of the application. BOD and COD concentrations decreased by 93% and 20% respectively. The total solid content of organic matter decreased by 94 percent due to microbial fragmentation (Anonymous, 2022a).

The Cassettesart University milk unit, established 36 years ago, is the first modern facility of its kind in the Kingdom of Thailand. 17 tons of day⁻¹ pasteurized and fermented milk are produced in the facility and 65 m³ wastewater is produced from the production. The BOD, COD and Solids Ratio of the resulting wastewater is 3319, 4767 and 1234 mg L⁻¹, respectively. Effective Microorganisms were used to treat the wastewater of this facility. EM, which is diluted with water at a ratio of 1:20, is sprayed twice a day on the surface of the pool. Within a few weeks, significant improvement was observed in the smell and crusting occurring on the surface.

When water was analysed, BOD was decreased 80% in the first pool and 94% in the second pool; COD was declined 79% in the first pool and 93% in the second pool; Solid matter decreased by 78% in the first pool and by 95% in the second pool. After continuous use of EM for 3 years, the situation in the pools was evaluated on site and laboratory-wide evaluation was carried out. In the plant, BOD averagely decreased 62% in the first pool and 94% in the second pool during the two-month period; COD decreased by 74% in the first pool and by 95% in the second pool. In laboratory-wide control and application reactor experiments, the benefit of EM was clearly seen in the reactor where EM is applied twice a day, the decrease in BOD and COD was 40-58% in the first pool and 60-75% in the second pool, respectively (Anonymous, 2022b).
The purpose of the use of EM is to transform an ecosystem contaminated and degraded by pathogenic microorganisms into an ecosystem containing productive and beneficial microorganisms. This simple principle is the basis of EM technology in agriculture and environmental management (Higa, 1993). The mixing of microorganisms into aquatic environments enriches the base of the ecological pyramid. When the bottom parts grow, the pyramid itself grows, resulting in richer diversity in the ecosystem. This diversity will help to increase the assimilation capacity of aquatic ecosystems, to be clean and beautiful again (Nikolopoulou and Kalogerakis, 2010).

Beytepe Pond, located in the Turkish National Botanic Garden Directorate (TMBB) campus in Ankara province, is used for irrigation and recreational purposes. The presence of flora and fauna in the Garden is closely related to the quantity and quality of the pond water. In this study, the availability of EM was investigated for improving the water quality of the Beytepe pond. For this purpose, water samples were taken from 6 different points between January and October 2021. Water temperature, pH, conductivity (EC), dissolved oxygen (DO), chemical oxygen demand (COD) and chlorophyll-a values were analysed from the samples. EM applications were made in the laboratory conditions in order to improve the quality of pond water.

2. MATERIALS AND METHODS

2.1 Research location and climate characteristics

Beytepe Pond (39°53 N, 32°45 E), which is located within the borders of the Turkish National Botanical Garden 15 km west of Ankara and was built in 1964 for irrigation and recreation purposes, has a surface area of 10.36 ha (Figure 1a). The pond is surrounded by woodland. The lake is a eutrophic lake, and the eutrophication increases over time. Factors affecting eutrophication and swamp formation in the Lake; they are materials of different origins that are transported to the lake with flood and precipitation waters and waste materials that come to the lake with groundwater from residential areas and industrial organizations. The pond is used as irrigation and for recreational purposes. The sustainability of the ecological and aesthetic benefits provided by Beytepe Pond is of great importance. The presence of TMBB natural areas under intensive urbanization in the centre of the city is made possible by the flora and fauna provided by Beytepe Pond.
The average climate data for many years in the field of study is presented in Table 1. The average annual temperature of the study area for many years is 11.9°C, the average sunbathing time is 6.8 hours, the total annual average precipitation is 391.9 mm.

Table 1. Average climate data for many years

| Mount | Tort (°C) | Tam (°C) | Tin (°C) | Sunbathing S. (hours) | Precipitation (mm) | Highest Temperature (°C) | Lowest Temperature (°C) |
|-------|----------|----------|----------|-----------------------|--------------------|-------------------------|--------------------------|
| 1     | 0.1      | 4.1      | -3.2     | 2.6                   | 40.2               | 16.6                    | -24.9                    |
| 2     | 1.6      | 6.4      | -2.3     | 3.8                   | 35.1               | 21.3                    | -24.2                    |
| 3     | 5.7      | 11.4     | 0.6      | 5.1                   | 39.1               | 27.8                    | -19.2                    |
| 4     | 11.2     | 17.3     | 5.3      | 6.6                   | 42.5               | 31.6                    | -7.2                     |
| 5     | 16       | 22.3     | 9.6      | 8.5                   | 51.5               | 34.4                    | -1.6                     |
| 6     | 19.9     | 26.6     | 12.9     | 10                    | 34.4               | 37                      | 3.8                      |
| 7     | 23.3     | 30.2     | 15.8     | 11.3                  | 14.3               | 41                      | 4.5                      |
| 8     | 23.3     | 30.4     | 15.9     | 10.7                  | 12.7               | 40.4                    | 5.5                      |
| 9     | 18.8     | 25.9     | 11.7     | 9.2                   | 18.0               | 37.7                    | -1.5                     |
| 10    | 13.1     | 19.9     | 7        | 6.7                   | 27.7               | 33.3                    | -9.8                     |
| 11    | 7.2      | 13       | 2.5      | 4.5                   | 31.5               | 24.7                    | -17.5                    |
| 12    | 2.4      | 6.4      | -0.8     | 2.5                   | 44.9               | 20.4                    | -24.2                    |
| Annual| 11.9     | 17.8     | 6.3      | 6.8                   | 391.9              | 41                      | -24.9                    |

2.2 Sampling
Water samples were taken from the surface and base at 6 sampling points (entrance, middle and outlet) selected in the lake to determine the concentrations of pollutants in the pond water (Figure 1B). Composite samples taken from the pond (sludge taken from the base were added) were used in laboratory experiment. Baikal EM1 (EM) was used as treatment material. Baikal EM1 provides synergistic effects by combining beneficial microorganisms such as lactic acid bacteria, yeast and phototrophic bacteria that exist in nature. It is a human-friendly and environmentally safe research product. Activates microorganisms that live in soil and water. It maximizes their natural power. Baikal EM1 contains the organisms Lactobacillus case, Rhodopseudomonas palustris, Saccharomyces cerevisiae, Lactococcus lactis. The total number of living organisms is 1,106 kobo. mL⁻¹.

2.3 Research topics and applications
Laboratory studies were carried out to improve the quality of pond water in controlled conditions. For this purpose, experiment was designed according to the following principles.
- Aerobic and anaerobic system (2 topics),
- Completion of the fixed and reduced part (the reduced amount is measured every 2 days and the volume is completed to 10 litres (2 subjects)
- EM application was applied 3 doses 3 recurrences including 0.5 ml L⁻¹, 1 ml L⁻¹, 2 ml L⁻¹. In the study, applications were carried out using 36 containers with a volume of 10 L.

2.4 Water Analysis
Monthly samples taken from sampling points and pH values were measured with pH meter and electrical conductivity with EC meter and dissolved oxygen concentrations with oxygen meter respectively. These measurements were made using the AZ 86031 Combo Water Quality

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Measurement Kit. EC, pH, DO and COD analyses were performed during sampling time. In addition, sampling was performed for microbiological analyses in quarterly periods. Microbiological analyses were performed in laboratories of the Department of Medical Microbiology at Gazi University. Types and numbers of microorganisms isolated from water samples are defined in in the same laboratory with conventional microbiological methods (CFU/MI colony forming unit = number of bacteria colonies).

3. RESULTS AND DISCUSSION

3.1 Results

3.1.1 Temperature, pH and Electrical Conductivity Values

The results of the mean pH, EC, DO and COD values of the samples measured during the monthly periods are shown in Table 2. pH values were lowest as 7.1 on the middle of the pond in June, with the highest in August on the left coastline and as measured 8.34. The average pH values ranged from 7.92 to 8.05. Pond water is grade II water according to pH value based on the Quality Criteria of the Continental Surface Water Resources of the Water Pollution Control Regulation.

Electrical Conductivity values are lowest as 0.96 dS m\(^{-1}\) on the left coastline in May and the highest was measured in January as 1.64 dS m\(^{-1}\) at the rain collector. The average measurement values ranged from 1.00-1.33 dS m\(^{-1}\). Electrical conductivity of the Pond water is grade III according to the U.S. salinity laboratory salinity classification (USSL).

The lowest value of dissolved oxygen was in September and measured as 2.30 mg L\(^{-1}\) at the bottom of the midpoint of the pond, while the highest value measured as 7.90 mg L\(^{-1}\) at the rain collector on July. The average value varied between 6.65-7.18 mg L\(^{-1}\). Cases where the dissolved oxygen (DO) values are below 5 mg L\(^{-1}\) are indicators of eutrophication in the lakes and ponds.

The lowest Chlorophyll-a value was 0.054 mg L\(^{-1}\) in the sample 2 in June, whereas the highest value was measured as 2.118 mg L\(^{-1}\) in August from the bottom in the midpoint of the pond. Spectrophotometric method was used in chlorophyll-a analysis. The pond exceeds the eutrophication limit values in terms of chlorophyll-a value.

The chemical oxygen demand is the most important test parameter used to determine the degree of pollution in water and wastewater samples. The lowest value of chemical oxygen demand was determined as 13 mg L\(^{-1}\) in the water sample taken from the surface of midpoint in June, and the highest was 1072 mg L\(^{-1}\) at the bottom of the midpoint in August. The COD value of the sludge taken from the bottom of the pond was 6980 mg L\(^{-1}\). The pond exceeds the eutrophication limit values in terms of COD value. COD analyses are determined by photometer method by using kits.

| Simple Point | pH    | EC (dS m\(^{-1}\)) | DO (mg L\(^{-1}\)) | COD (mg L\(^{-1}\)) |
|-------------|-------|-------------------|-------------------|-------------------|
| 1           | 8.00  | 1.00              | 6.78              | 45.70             |
| 2           | 7.95  | 1.04              | 6.65              | 42.80             |
| 3           | 7.99  | 1.33              | 6.81              | 17.00             |
| 4           | 8.05  | 1.13              | 0.00              | -                 |
| 5           | 8.00  | 1.02              | 7.18              | 16.15             |
| 6           | 7.92  | 1.01              | 7.03              | 15.70             |

Types and numbers of microorganisms isolated from water samples Coliform bacteria are defined in in laboratories of the Department of Medical Microbiology at Gazi University with conventional microbiological methods (CFU/MI colony unit = number of bacteria colonies).

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3.1.2 The Chemical Oxygen Demand

The COD value of raw water used as a result of the composite sample and sludge mixture used at the beginning of laboratory applications is 263 mg L\(^{-1}\). COD values were measured twice as in the 10th day and in the end of the application (Day 20) (Figure 2). The lowest COD value was determined as 2 mg L\(^{-1}\) in the closed system with 0.5 ml L\(^{-1}\) EM application and constant amount of water. The highest value was determined as 46 mg L\(^{-1}\) with 2 ml L\(^{-1}\) EM application in the open system-ventilation and constant amount of water.

In the treatment studies, the initial COD value was above the eutrophication limit value and the appropriate value has been reached in the application with the 0.5 ml L\(^{-1}\) EM at the closed system and with the constant water.

![Figure 2. COD (mg L\(^{-1}\)) Values During the Application](image)

3.1.3 The Dissolved Oxygen

The DO value was measured at 2.4 mg L\(^{-1}\) at the beginning of the treatment and increased to minimum 4.93 mg L\(^{-1}\) at 2 ml L\(^{-1}\) EM application in closed system with constant water and reached to maximum value as 7.4 mg L\(^{-1}\) in the open system with 0.5 and 2 ml L\(^{-1}\) EM application and

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continuous flow. Dissolved oxygen values were measured on day 8 and day 16 on day 4 during application (Figure 3).

3.1.4 Chlorophyll -a

The raw water supply must be maintained to slow the increase in the concentration of nutrient substances that will occur over time in surface waters. Chlorophyll analyses are carried out in order to determine the productivity and biomass in the water sources. Composite sample chlorophyll-a value was measured as 1.54 mg L\(^{-1}\) at the beginning of the study.

As a result of the treatment studies, chlorophyll-a values were reached to the minimum value as 0.2 mg L\(^{-1}\) at the closed system with constant water with 0.5 ml L\(^{-1}\) Effective Microorganism application. Maximum value was 0.247 mg L\(^{-1}\) at the open system with 0.5 ml L\(^{-1}\) Effective Microorganism application (Figure 4). Although the applications improved the chlorophyll-a value, the eutrophication limit value was not reached.

![Figure 3. DO (mg L\(^{-1}\)) Values During the Application](image)

3.2. Discussion

With the application of effective microorganism technology in septic systems, COD and BOD concentrations decreased in lagoons, active sludge systems and other healing processes. EM applications greatly reduce the smell of pig manure and coliform bacteria (Rashid and Wesd 2007).

In a study, Namsivayam et al. (2011) evaluated the EM formulation in terms of alkalinity under standard conditions, total dissolved solids, biological and chemical oxygen demand. All the tested parameters showed a marked reduction. The result of the test indicates that EM has the potential to increase the treatment effectiveness of domestic wastes.

A study in Sungai Kelian, Perak, Malaysia, found that EM mud balls improved the river's water quality and cleared mud from the river six months after the mud balls were thrown. Suspended solids, dissolved oxygen, COD, BOD, ammonia nitrogen and pH were analysed in the samples.

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taken from the river before the application; and water quality was determined to be Class IV for irrigation. It is stated that water quality improved to class III after the application (Zakary et al. 2010). Similar to the results, 67% and 99% improvement was determined in the values of DO and COD, respectively in the present study.

Since the nutrient load is an important parameter for eutrophication, effective microorganisms (EM) were used in the treatment of industrial wastewater from the Bilqas sugar beet plant in Dhaahliya province, Egypt, to investigate the effect of organic loading speed on removal. Bilqas Sugar Factory wastewater treatment plant is divided into two main stages. In the first stage, the wastewater used in beet washing is separated by mechanical separator, clean water is reused for beet washing and the sludge is removed from the wastewater. The second phase is the active sludge process. EM has been applied in both phases. COD\BOD\TSS\TDS values were 1703.7\1166.5\1106.6\1600.1 mg L\(^{-1}\), while 129.8\88.1\150.4\1460.0 mg L\(^{-1}\) was observed in the output water. The potential for odour removal and sludge reduction is quite good. In addition, active sludge operating costs decreased (Embaby et al. 2010). Similarly, EM applications showed a 77% improvement in chlorophyll-a values, which are eutrophication markers. As a result of the treatment with EM, the odour problem has also been eliminated.

4. CONCLUSION
Effective Microorganisms (EM) consist mainly of lactic acid bacteria, photosynthetic bacteria, yeast, fermentation fungi and actinomycetes. It is a culture in which beneficial microorganisms coexist, increasing plant growth and yield by increasing microbial turnover in the soil. The use of EM in the fields of agriculture, livestock, environment and human health has become extremely widespread all over the world in the last 20 years. It is also used for the treatment of sewage or wastewater.

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Effective Microorganisms are known to improve water quality parameters such as turbidity, dissolved oxygen and pH in environmental applications. In the applications in water samples taken from Beytepe Pond, effective microorganisms have been effective on dissolved oxygen and pH values, but have also provided close improvements in chemical oxygen demand and the chlorophyll-a values.

As a result of the study, it was concluded that 0.5 mL L⁻¹ application to Beytepe Pond water can be applied without any ventilation process. Effective microorganisms provide a cleaner and healthier environment without environmental impacts. They reduce the presence of pathogenic microorganisms and the use of chemicals in water and soil. With these properties, its use as an environmentally friendly material should be widened.

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