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RESEARCH ARTICLE

ISOLATION OF MICROALGAE FROM ANTARCTIC SOIL

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ABSTRACT

Microalgae are unicellular photosynthetic microorganisms which are able to survive in extreme environmental conditions, specifically low temperature, high evaporation rates as well as low water availability. Polar microalgae from the Antarctic region have received attention from researchers due to their special adaptations for surviving in extremely cold environments and being isolated from various types of samples. Four Antarctic soil samples were used on this study namely-Sample A, Sample B, S18 and S30. All soil samples were subjected to serial dilution and spread on BBM followed by 3 weeks incubation at 25°C with periodic light. Only soil sample S30 showed positive microalgae growth after additional 3 weeks incubation period. Consequently, soil sample S30 was proceeded to isolation of microalgae using BBM, 3N-BBM+V, JM and MWC media. Observable growth of green microalgae could only be obtained using 3N-BBM+V and JM media. Moreover, 3N-BBM+V medium successfully produced the highest number of microalgae colonies recovered from Antarctic soil sample. Isolation of microalgae from Antarctic provides resources for various industrial and commercial applications.

KEYWORDS

Antarctic Soil, Isolation, Microalgae, Polar Region, Spread Plate

1. INTRODUCTION

The Antarctic, which is almost covered completely by an ice sheet, actually possesses about 2% of ice-free region. It is also known as the coldest, windiest as well as driest continental region on the Earth. Despite of the presence of these conditions, many Antarctic microorganisms have been detected in various forms of habitats which include ponds, lakes, rivers, rocks and even soil (Nunez-Montero and Barrientos, 2018). Common factors such as the existence of vegetation, the physicochemical parameters of the region and altitude are influencing the diversity of the Antarctic microbes (Malcheva et al., 2020). Many scientists are interested in investigating the special adaptation strategies of these microorganisms which could help them to survive in those harsh environments. Knowledge gained from those studies can be used to identify the potential of these microbes in meeting the current challenges in various sectors including industrial, medical and commercial (Lambrechts et al., 2019). For example, Antarctic microbes have been identified as the new potential source for producing antimicrobial compounds to facilitate the emergence of antibiotic-resistant bacteria which compromise the production of pharmaceutical drugs, food as well as agricultural products (Nunez-Montero and Barrientos, 2018). Photoprotectants produced by microalgae due to solar radiation can be used for the development of suncare products against photoaging (Rastogi et al., 2017). Antarctic microalgae also have been recognized as the new candidate for anti-cancer agents in fighting human cancer since they have the ability to induce apoptotic cell death in cancer cells and exhibit antioxidant activity (Suh et al., 2017).

Microalgae constitute a varied microbial community from the Antarctic and have been recognized for their ability to survive in extreme environments particularly at very low sub-zero temperatures (Orekhova et al., 2018). A number of unicellular microalgae have been identified and reported from the past research conducted at Antarctic. Some of the microalgae species which have been found in the Antarctic region include *Koliella antarctica*, *Chlorella sp.*, *Chlamydomonas sp.*, *Chromulina spp.*, *Sanguina spp.*, *Chloromonas spp.* and *Coccomyxa subellipsoidea* (La Rocca et al., 2015; Rivas et al., 2016; Liu et al., 2016; Soto et al., 2020; Pfaff et al., 2016). Additionally, microalgae play an important role in Antarctic environments as nutrients providers for other living organisms. Some of the mechanisms which could help them to survive in low temperature include synthesizing high levels of sugar and polyols, producing anti-freeze proteins as well as altering lipid content (Orekhova et al., 2018). Many studies have been conducted by researchers to isolate microalgae from various types of samples originating from the Antarctic region such as snow surface as well as biological soil crusts (BSCs) (Cid-Aguero et al., 2017; Borchhardt and Grundling-Pfaff, 2020). However, the use of solely soil samples from the Antarctic in microbiological study is very limited and need to be further investigated in order to isolate and identify the species of microalgae which can survive in the cold environment. Hence, this study was carried out by using Antarctic soil as a sample for the isolation of microalgae.

2. MATERIAL AND METHODS

2.1 Materials

Soil samples collected from the Antarctic by Dr. Noor Faizul Hadry from International Institute for Halal Research and Training (INHART)

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International Islamic University Malaysia were used in this study and were stored at -20°C. The Antarctic soil samples were labelled as follows: soil sample A, B, S18 and S30.

2.2 Isolation of Microalgae Using Bold Basal Medium (BBM)

About 1 g of soil sample was added into 100 ml BBM broth followed by incubation at 23 - 25°C for 1 week with periodic light and shaking at 120 rpm. Next, all soil samples were subjected to serial dilution of 10^{-1} until 10^{-5}. About 100 µl of sample from each dilution was pipetted and spread on BBM medium in duplicates. All the plates were incubated in an incubation room at 23 - 25°C for 3 weeks with periodic light.

2.3 Isolation of Microalgae from Soil Sample S30 Using BBM, 3N-BBM+V, Modified Wright's Cryptophyte (MWC) and Jaworski's Medium (JM) Media

Serial dilution of 10^{-1} until 10^{-3} was performed on soil sample S30 and 100 µl of sample for each dilution was pipetted and spread on BBM, 3N-BBM+V, MWC and JM media in duplicates. All the plates were incubated in an incubation room at 23 - 25°C for 3 weeks with periodic light.

2.4 Morphology Observation

Colonies of microalgae were physically characterized by observing their colour. Number of microalgae isolated also were being noted.

3. Results and Discussion

Four Antarctic soil samples, namely sample A, sample B, sample S18 and sample S30 were used in this study. After three weeks of incubation period on BBM medium, no growth of microalgae was detected for all samples with undiluted and diluted samples (10^{-1}-10^{-5}). There was no observable growth of microalgae on any of the cultured BBM agar plates due to several possible factors. Microalgal cells may still in the resting stage. Resting stage is a common strategy used by polar microalgae to remain survive in extreme environments where it will initiate the formation of resting cells, spores or cysts. Their metabolic reactions in producing biomass will slow down since the production of resting stage cells becomes their main focus. Besides this strategy has a significant role in conserving microalgal species biodiversity, it also could provide energy storage which later can be used by microalgae for their survival (Orlavo and Morozowa, 2009). A group researchers reported that upon the encounter of stressful conditions, microalgae would enter a resting stage and similar cases were observed in a study done (Pushkareva et al., 2016; Pichrtoya et al., 2014). However, microalgae growth can be initiated again if essential requirements or conditions are being met. Since none of microalgae were grown on the BBM plates in this study, maybe not many of them remained viable and needed a longer incubation time so that a visible growth can be observed.

Furthermore, these soil samples were stored for a quite some time in a -20°C freezer before being used for microalgal isolation purposes. Prolonged storage at very low temperature might reduce the recovery levels of microalgal cells due to ice formation and salt induced injuries. Based on a study done, microalgal cultures which have been stored at -15°C for one month have lost viability rapidly as determined by the pour-plate assay (Kapoore et al., 2019). They also showed a slow growth pattern in a nutrient-rich environment which suggested that microalgae might require some time for reviving again due to their adaptation phase for adjusting in the new conditions. Since no microalgal growth was obtained after the incubation period of three weeks, plates were further incubated for another three weeks to check whether a longer incubation period is required to promote microalgal growth. From the results, only soil sample S30 showed visible green growth of putative microalgal on the wall of conical flask as indicated in Figure 1. Sample S30 was then proceeded for further isolation of microalgae by using another three different media namely, 3N-BBM+V, MWC and JM as an attempt to obtain different types and strains of microalgae.

From the observation, microalgae were successfully isolated from soil sample S30 using 3N- BBM+V and JM media as depicted in Figure 2 and Figure 3. MWC medium, however, failed to produce any microalgal colonies except for fungus. All colonies are green and the range of colony number obtained on 3N- BBM+V was < 600 – 41 compared to < 300 – 37 on JM medium for 10^{-1} to 10^{-3} dilution factor. Higher number of microalgal growth was obtained using 3N-BBM+V as the culture medium is probably because the medium has a rich nutrient composition and could facilitate the growth of microalgae more readily. The observed colonies were green in colour and most likely identified as microalgae since the media used were specifically would promote the growth of these microorganisms.

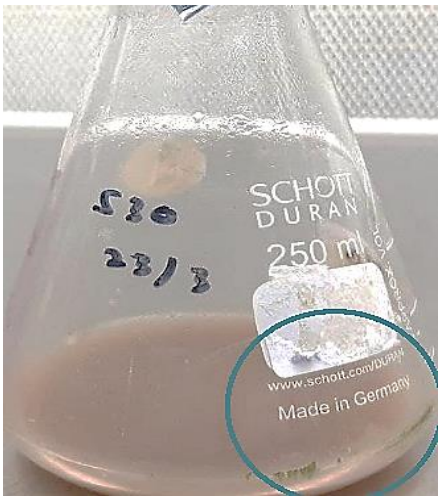

Figure 1: Visible green growth of putative microalgal in sample S30

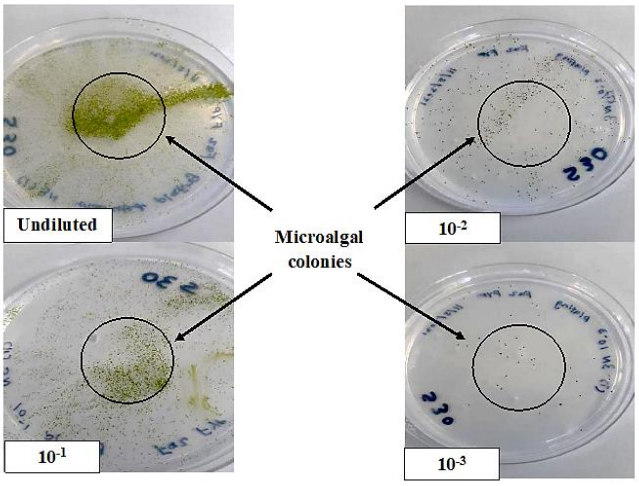

Figure 2: Microalgal colonies isolated from sample S30 on 3N-BBM+V medium

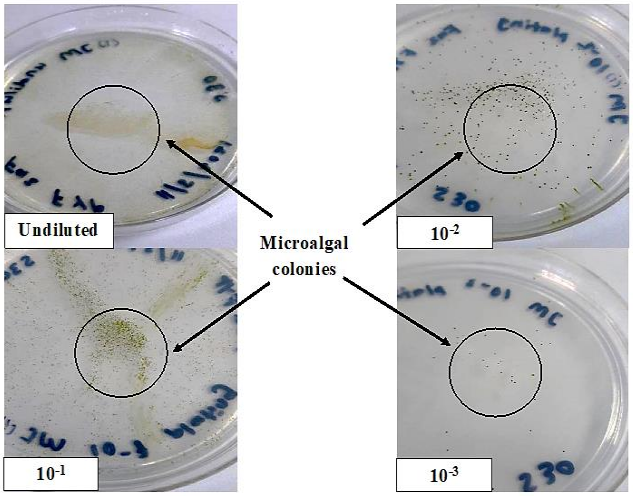

Figure 3: Microalgal colonies isolated from sample S30 on JM medium

4. Conclusion

In conclusion, microalgae are eukaryotic microorganisms which could survive in various types of environments including the extremely low temperature of the Antarctic region. Four different Antarctic soil samples (Soil A, Soil B, S18, S30) were used to isolate microalgae using BBM, 3N-BBM+V, JM and MWC as the culture media. Based on findings, microalgae was successfully isolated from soil sample S30 and 3N-BBM+V agar plates had the highest number of microalgal colonies as compared to JM plates. It can be deduced that 3N- BBM+V medium is the best candidate for culturing microalgae.

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REFERENCES

Borchhardt, N., Gründling-Pfaff, S., 2020. Ecophysiological Response Against Temperature in Klebsormidium (Streptophyta) Strains Isolated From Biological Soil Crusts of Arctic and Antarctica Indicate Survival During Global Warming. Frontiers in Ecology and Evolution, 8(June), Pp. 1–10.

Cid-Agüero, P., Cuello, J.L., Ruiz, S., Sanchez, G., 2017. Growth and lipid profiles of the Antarctic snow microalga Chlamydomonas sp. In response to changes in temperature, photoperiod, salinity and substrate. Anales Del Instituto de La Patagonia, 45 (3), Pp. 45–58.

Kapoore, R.V., Huete-Ortega, M., Day, J.G., Okurowska, K., Slocombe, S.P., Stanley, M.S., & Vaidyanathan, S., 2019. Effects of cryopreservation on viability and functional stability of an industrially relevant alga. Scientific Reports, 9 (1), Pp. 1–12.

La Rocca, N., Sciuto, K., Meneghesso, A., Moro, I., Rascio, N., Morosinotto, T., 2015. Photosynthesis in extreme environments: Responses to different light regimes in the Antarctic alga Koliella antarctica. Physiologia Plantarum, 153 (4), Pp. 654–667.

Lambrechts, S., Willems, A., Tahon, G., 2019. Uncovering the uncultivated majority in antarctic soils: Toward a synergistic approach. Frontiers in Microbiology, 10(FEB), Pp. 1–19.

Liu, C., Wang, X., Wang, X., Sun, C., 2016. Acclimation of Antarctic Chlamydomonas to the sea-ice environment: a transcriptomic analysis. Extremophiles, 20 (4), Pp. 437–450.

Malcheva, B., Nustorova, M., Zhiyanski, M., Sokolovska, M., Yaneva, R., Abakumov, E., 2020. Diversity and activity of microorganisms in antarctic polar soils. One Ecosystem, 5, Pp. 1–16.

Núñez-Montero, K., & Barrientos, L. (2018). Advances in antarctic research for antimicrobial discovery: A comprehensive narrative review of bacteria from antarctic environments as potential sources of novel antibiotic compounds against human pathogens and microorganisms of industrial importance. Antibiotics, 7 (4).

Orekhova, A., Barták, M., Hájek, J., 2018. Post rapid freezing growth of Antarctic strain of Heterococcus sp. monitored by cell viability and chlorophyll fluorescence. Cryobiology, 85, Pp. 39–46.

Orlova, T.Y., Morozova, T.V., 2009. Resting stages of microalgae in recent marine sediments of Peter the Great Bay, Sea of Japan. Russian Journal of Marine Biology, 35 (4), Pp. 313–322.

Pfaff, S., Borchhardt, N., Boy, J., Karsten, U., Gustavs, L., 2016. Desiccation tolerance and growth-temperature requirements of Coccomyxa (Trebouxiophyceae, Chlorophyta) strains from Antarctic biological soil crusts. Algological Studies, 151–152 (1), Pp. 3–19.

Pichrtová, M., Hájek, T., Elster, J., 2014. Osmotic stress and recovery in field populations of Zygnema sp. (Zygnematophyceae, Streptophyta) on Svalbard (High Arctic) subjected to natural desiccation. FEMS Microbiology Ecology, 89 (2), Pp. 270–280.

Pushkareva, E., Johansen, J.R., Elster, J., 2016. A review of the ecology, ecophysiology and biodiversity of microalgae in Arctic soil crusts. Polar Biology, 39 (12), Pp. 2227–2240.

Rastogi, R.P., Sonani, R.R., Madamwar, D., 2017. UV Photoprotectants From Algae-Synthesis and Bio-Functionalities. Algal Green Chemistry: Recent Progress in Biotechnology, Pp. 17–38.

Rivas, C., Navarro, N., Huovinen, P., Gómez, I., 2016. Photosynthetic UV stress tolerance of the Antarctic snow alga Chlorella sp. modified by enhanced temperature? Revista Chilena de Historia Natural, 89, Pp. 1–9.

Soto, D.F., Fuentes, R., Huovinen, P., Gómez, I., 2020. Microbial composition and photosynthesis in Antarctic snow algae communities: Integrating metabarcoding and pulse amplitude modulation fluorometry. Algal Research, 45, Pp. 101738.

Suh, S.S., Yang, E.J., Lee, S.G., Youn, U.J., Han, S.J., Kim, I.C., Kim, S., 2017. Bioactivities of ethanol extract from the antarctic freshwater microalga, chloromonas sp. International Journal of Medical Sciences, 14 (6), Pp. 560–569.