# UNIVERSITI TEKNOLOGI MARA

# EFFECT OF *Aspergillus brasiliensis* AND *Sphingobacterium spiritovorum* INTERACTION ON THE KINETIC REMOVAL OF BENZO(A)PYRENE IN WASTEWATER

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# ABSTRACT

Wastewater from the petrochemical industry commonly contains xenobiotic high-molecular-weight polycyclic aromatic hydrocarbons (HMW-PAHs). Prior to utilising the resources for human use, it is crucial to eliminate the pollutants in the aquatic environment, considering their potential risks to human health. Several bioremediation studies have been implemented. However, the investigation of the interaction between bacteria and fungi has been relatively limited. *A. brasiliensis* and *S. spiritovorum* were used to evaluate the potential interaction to degrade benzo(a)pyrene in wastewater as in monoculture and co-culture systems. The study begins with optimization of initial pH and temperature by Design-Expert software. The kinetic response of monoculture and co-culture (simultaneous and sequence) was evaluated with an in-depth analysis of the co-culture. This includes analysing the interaction type, surface hydrophobicity and adhesion prediction by using The Lotka-Volterra model, contact angle measurement (CAM) and thermodynamic DLVO theory respectively. A mathematical modelling study was conducted to analyse the growth kinetics of monoculture and co-culture while degrading BaP. The Design Expert examination indicated that *A. brasiliensis* thrives at a pH of 5.5 and a temperature of 40 ℃, while *S. spiritovorum* performs best at a pH of 7.6 and a temperature of 25 ℃. While co-culture is best at an initial pH of 6.0 and a temperature of 25 ℃. The efficacy of Design-Expert in optimising BaP degradation was confirmed using ANOVA analysis. Significantly, sequential treatment surpassed simultaneous, and monoculture treatment, exhibiting greater and faster removal of BaP of 79 % at 10 mg/L and more than 50 % for other concentrations. The study demonstrated the dominance of *A. brasiliensis* in the microbial community, its role in BaP degradation, and its symbiotic connections with *S. spiritovorum*. By altering the physicochemical properties, the degradation of BaP in co-culture was increased. The study of adhesion dynamics provided a clear understanding of the complex interaction between microorganisms, highlighting the significant influence of surface hydrophobicity. The Monod model was accurately applied to the correlation between the specific growth rate and substrate concentration derived from the growth curves. The kinetic parameter of an inhibitory model proposed by Haldane was effectively determined using the experimental data. The fourth-order Runge-Kutta method was extensively enhanced to compute the optimal parameter for a given non-linear problem. These findings provide insights into the interactions between *A. brasiliensis* and *S. spiritovorum*, highlighting their physicochemical characteristics and the breakdown of BaP. The mathematical modelling derived from these interactions can guide the development of more efficient bioremediation strategies for polluted environments, improving the degradation of contaminants in wastewater and soil treatment systems.

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# INTRODUCTION

## 1.1 Research Background

The rapid growth of the exploiting, utilizing and refining of crude oil prior to energy demand and oil recovery technology had caused many incidents of oil spills in the water supply. Polycyclic Aromatic Hydrocarbons (PAHs) are one of the toxic and carcinogenic pollutants from incomplete combustion of crude oil which require to be eliminated from the environment. In Malaysia, the main contributor to the presence of PAHs in an aquatic environment is the used crankcase oil. The PAH presence is through spillage and dumping of the waste directly into the water bodies and street runoff which leakage from vehicles (Zakaria, 2002; Wang et al., 2022). Places near to harbour including Langkawi Island (Nasher et al., 2013; Terence and Masni, 2021), Penang (Sakari and Zakaria, 2013), Klang Valley (Bakhtiari et al., 2010) and Malacca Straits (Manan et al., 2011) experienced an excessive amount of PAHs concentration. The PAHs are distributed once enter water bodies to the river or marine sediments which eventually accumulate in aquatic life such as lobster, muscle and clams through absorption (Abdel-Shafy and Mansour, 2016). This has become a persistent pressure on the environment and human beings due to its fused ring compounds that are less prone to oxidative damage (Vasudevan et al., 2018). The United States Environmental Protection Agency (US EPA) has listed 16 PAHs as high-priority pollutants and benzo(a)pyrene has been recognized as a potential carcinogenic pollutant by the World Health Organization (World Health Organization, 2017). Numerous approaches have been developed to minimize the toxicity of PAHs in the past two decades including physical, chemical and biological methods. Owing to its environmentally friendly and cost-effective features, the bioremediation technique is by far the most popular field of study compared to both chemical and physical methods of remediation.

However, the high hydrophobic carbon chain of PAHs is widely accepted as limits the bioavailability of the pollutants to the bacteria and slows the biodegradation rate (Folwell et al., 2016). Many approaches have been studied to enhance the biodegradation of the PAHs including identifying the best type of bacteria, and controlling environmental factors such as pH, temperature, oxygen content and